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Rediscovering the Maryland Darter (Etheostoma sellare)

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Rediscovering the Maryland Darter (*Etheostoma sellare*)

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Rediscovering the Maryland Darter (*Etheostoma sellare*)

Tyler Russell Hern

The Maryland darter has not been observed since 1988. Historic populations were located in Deer Creek, Swan Creek, and Gashey's Run all of which occur in the lower Susquehanna drainage. At these locations, specimens were collected or observed at the lowest riffle of the stream. Some researchers suggest this may be a large river darter. The Conowingo Dam complicates surveying below the facility due to the rapid fluctuations in water levels caused by regulation for power production. Surveying efforts included trawling in the mainstem, and visual surveys in the mainstem, and searching/sampling all historically known locations of *Etheostoma sellare*. During this study 153 Benthic trawls totaling 272.4 minutes of bottom time yielded no Maryland darters. Also 10,452 fish were collected from 4 tributary sites (3 historic and 1 new) yielding no Maryland Darters. In addition, during 307.1 man-hours of visual surveys no Maryland darters were observed. Considering the data from this study it is apparent that the Maryland darter has declined within its historical range. However, habitat assessment of new sites showed that habitat similar to *E. sellare's* historical habitat still exists in the Susquehanna mainstem and tributaries. Additional surveys are needed to determine if the Maryland darter is extant.

1.0 Introduction

1.1 Historical Context

About 135 million years ago during the late-Triassic period, the super-continent of Pangaea began to separate through the processes of plate tectonics. These movements greatly influenced the distribution of world biota, especially fishes. Separation due to plate movement has caused some species/families to exist only in certain areas. For example, lungfishes (Lepidosirenidae) occupy regions that derived from Gondwanaland, while paddlefishes (Polyodontidae) occupy regions derived from Laurasia (Helfman et al. 1997). Plate tectonics also influenced fish distributions through the formation of mountains. The rise of the Appalachian Mountains played a major role in the present-day arrangement of fishes in the northeastern United States (Hocutt et al., 1986). This physical mountain barrier most likely prevented the colonization of upland watershed, streams in the Appalachian Plateau or Piedmont Physiographic Province of Maryland, by some species.

At the end of the last ice age, ocean levels rose and created additional migration barriers. The rising seas flooded areas that were formerly the mouths of streams, forcing fluvial species farther upstream.

These natural phenomena are responsible for creating the unique habitat that occurs along the coastal plain region of Maryland and Virginia.

Combinations of plate tectonics, geographic barriers and natural phenomena has created a habitat and fish community in the upper Chesapeake Bay area that is found nowhere else on Earth. Numerous species, including the Maryland darter (*Etheostoma sellare*), have adapted to this area and their survival is dependent on the conservation of this unique ecosystem.

1.2 Human Impacts on the Susquehanna River

The lower Susquehanna River Watershed is one of the first areas settled in America by Europeans. The streams in the area has seen drastic changes due to human influences dating back to the early 1600s. When European settlers first arrived in the Lower Susquehanna River Basin, streams meandered through unbroken forests and wetlands (Stranahan, 1993). Eventually, farmers cleared forests for crops and pasture lands. This deforestation resulted in habitat degradation though sedimentation, threatening aquatic ecosystems.

The early settlers brought the first changes to the Lower Susquehanna River basin. As the human population in the area grew certain species of fish were extensively harvested, and fish numbers plummeted. The American shad (*Alosa sapidissima*) and blueback herring (*Alosa aestivalis*) were nearly extirpated due to the unregulated fishing. The timber industry made its mark on the Susquehanna River when it boomed in the late 1800s. Thousands of acres of forest were clear-cut, and the river being the most practical way for timber to be moved, was filled with logs as they floated downstream to mills for processing. Clearing trees within the watershed made streams warmer, muddier, and more susceptible to erosion. The effects of the timber boom were felt throughout the Susquehanna watershed further degrading the once pristine waters.

More recently the lower Susquehanna River basin has been affected by increasing urbanization. The Coastal Plains region of Maryland has the highest rate of urbanization in the state (Roth et al., 1999). In association with urbanization comes increasing amounts of impervious surface area through construction of homes, parking lots, and roads. Impervious surface increases rates of runoff, thus leading to higher stream velocities and erosion. Recent studies have shown that when as little as 1% of a watershed becomes impervious certain intolerant species (e.g. Brook trout) can disappear from that watershed (Stranko & Klauda, 2010). Along with urbanization comes the need to supply the growing population with utilities: electricity, drinking water, wastewater treatment, solid waste disposal, and many others, all of which have potentially detrimental effects on the natural environment. The current rate of population growth and development will cause urbanization to expand to an area the size of Baltimore County, MD (612 sq. mi.) over the next 25 years (Boward et al., 1999). Models also suggest that by the year 2030, 296 streams in Maryland will become unsuitable for species of concern in Maryland, and 75 percent of those streams are located in the Coastal Plain region (Hern, 2006).

All of these anthropogenic factors greatly alter the assemblages of stream biota. Impacts like these cause problems for species, such as darters, that rely on clean substrate for habitat and spawning beds. Darters also rely on benthic macroinvertebrates and gastropods for food; when sediment covers the habitat these organisms attach to for survival, resources become limited and survival of these species is jeopardized.

1.3 Effects of Dams

Damming includes the construction of physical barriers like impoundments, flood control dams, and hydroelectric facilities and their effects have been well documented (Saunders and Hobbs, 1991; Dynesis & Nilsson, 1994; Pringle et al., 2000; Tiemann et al., 2004; Layman et al., 1993; Mattingly & Galat, 2002; Travnichek et al., 1995; Morita & Yamamoto, 2002; Bradford, 1997; Irvine et al., 2009; Saltveit et al., 2001; Garcia et al., 2009). Dams are a major threat to aquatic ecosystems around the world (Saunders & Hobbs, 1991; Dynesis and Nilsson, 1994; Pringle et al., 2000; Tiemann et al., 2004; Morita et al., 2000). Dams prevent movement of species and nutrients through fragmentation, alter flow regimes, and directly endanger aquatic organisms through the possibility of stranding (Bradford, 1997, Irvine et al, 2009; Saltveit, 2001).

Fragmentation of watersheds disrupts the continuous nature of aquatic ecosystems. Studies have shown the importance of connectivity, between headwater streams and downstream rivers, and bays for the persistence of populations (Morita and Yamamoto 2002). By interrupting the flow of nutrients, dams can potentially alter the species richness and diversity of downstream ecosystems. Fragmentation of aquatic habitats increases the rate of extinction (Wilcox & Murphy, 1985). Extirpations of some freshwater fishes directly link to the fragmentation of watersheds by damming (Winston et al., 1991).

Besides the physical barriers created by dams, they also drastically alter stream flow rates. Flow velocities are especially important because they determine the rate at which nutrients and oxygen reach aquatic organisms. Some fishes require minimum flow velocities in order for areas to remain suitable for habitation. When dams change or regulate flow velocities this minimum viable flow is jeopardized.

Migratory fish have been severely impacted by human activities, especially dam building. Historically canal dams across the Susquehanna River restricted access to upstream spawning and nursery grounds. Now, the large hydroelectric dams on the lower Susquehanna River completely eliminated access to all but the lower 10 miles of the river. As a result, up-running migratory fish can no longer reach spawning and rearing habitat in the upper Susquehanna River without passing through these facilities. American shad, alewife (*Alosa pseudoharengus*), blueback herring, striped bass (*Morone saxatilis*), Atlantic sturgeon (*Acipenser oxyrinchus*), shortnose sturgeon (*Acipenser brevirostrum*), and American eel (Anguilla rostrata) are species whose migrations have been interrupted by impoundments on the Susquehanna River.

Hydroelectric facilities cause artificial fluctuations in stream flows when water is held or released for aid in power production. These artificial fluctuations can degrade fish habitat and create a situation unsuitable for many fluvial fish species. Studies have shown that regulating minimum flow can increase fluvial species diversity up to 40% within 3 kilometers of the facility. An increase in fluvial species was detected as far as 37 kilometers downstream (Travnichek et al., 1995).

The introduction of invasive species into an ecosystem can have drastic effects on the native organisms. Reservoirs resulting from stream impoundments are often stocked with non-native game fishes. Species such as largemouth bass (*Micropterus salmoides*) and bluegill (*Lepomis macrochirus*) are extensively stocked into reservoirs for sport fishing. These species often cause declines in native freshwater fishes by outcompeting for available resources (McKinney 2001). Maryland hosts at least 14 invasive aquatic organisms, including the northern snakehead (*Channa argus*) and grass carp (*Ctenopharyngodon idella*) (McKinght, 2011). Nearly all introductions of nonnative species have had detrimental effects on native stream biota. Introduced species are at least partially responsible for 24 of the 30 freshwater species that have become extinct, in the US, in the last 100 years. In addition 2 out of 30 of these extirpated species have become extinct primarily because of the direct or indirect effects of introduced species (Miller et al., 1989).

An often overlooked effect that dams can have on local fish assemblages is the increased risk of stranding. When hydroelectric dams regulate for power and cause rapid decreases in flow there is potential for fish to become trapped on gravel and sand bars. Gravel and sand bars are sometimes exposed when water levels drop as a result of this regulation. Recent studies have shown that some fish are trapped in side-channels and on exposed land as water levels drop (Bradford, 1997; Irvine et al., 2009; Saltveit et al., 2001). Conversely, Irvine et al. (2009) suggested that the rate of dewatering by dams was not significant in the incident of stranding. However, they did show that the number of fish that were trapped in side-channels was positively correlated with the rate of dewatering.

Studies have shown stream impoundment specifically lead to the decline of darter species (Wine et al., 2008; Layman et al.1993, and Mattingly & Galat, 2002). Downstream effects such as fragmentation of watersheds, interruption of fish migrations, and reduction of habitat connectivity are responsible for these declines (Garcia et al., 2009; Tiemenn et al., 2004). However, upstream effects may be equally responsible for declines due to changing water depths, flow velocities, and hydrologic processes (Winston et al., 2001).

The sheer number of blockages on Maryland's waterways is staggering and one blockage in particular has a direct effect on Maryland darter habitat: The Conowingo Hydroelectric Project that was completed in 1928. This massive hydroelectric operation is located 3.8 kilometers upstream from the last known location of the Maryland darter population.

1.4 Darter Ecology

Darters are members of the Percidae family. They are unique among North American fishes because they only occur east of the Rocky Mountains, which suggests that they immigrated or evolved in North American relatively recently in geologic terms. The Percidae family contains 10 genera and about 200 species in North America, a relatively large number of species surpassed in diversity only by the minnow family (Cyprinidae). Most of the Percidae species are small, bottom-dwelling fishes that move quickly from beneath rocks, which is how they got their name “darters”. Other genera in the Percidae family in North America (Genera *Perca* (perch) and *Sander* (walleye and sauger) are large in comparison to the darters.

Etheostoma is the largest and most diverse genus of darters. Kuehne and Barbour (1983) list 94 distinct species, but recognize that the genus may grow closer to 120 species as species complexes are separated. Within the genus *Etheostoma* there are 17 subgenera *(Psychromaster, Litocara, Allohistium, Etheostoma, Ulocentra, Doration, Boleosoma, Ioa, Valiiantia, Nothonotus, Villora, Ozarka, Austroperca, Oligocephalus, Catonotus, Hololepis, and Microperca)* (Kuehne & Barbour, 1983). The subgenus *Etheostoma* is comprised of 14 species that share a combination of the following characteristics: eyes set high on the broad head with a rounded snout; heavy lips; usually a well developed frenum; gill membranes broadly joined; large round pectoral fins; supratemporal and infraorbital sensory canals complete; first anal fin spine thick and stiff; genital papilla of females a long rounded tube; and genital papilla of males short flattened tube. Males are larger than females in many species. This subgenera is further divided into three species groups (*E. variatum, E. thalassinum, and E. blennioides*) (Richards, 1966).

Darters range in area from the Hudson Bay to the Gulf of Mexico and inhabit a large range of habitats. Certain species of darters are widespread (*E. olmstedi* and *E. nigrum*) and inhabit multiple habitat types, while others can be found only in specific locations (*E. nuchale* and *E. sellare*).

Unlike their larger Percidae relatives walleyes and saugers, darters select specific spawning sites and many species provide parental care for the eggs and fry, which increases survival rate. Darters do not release their entire seasonal egg production at one time; instead, they release a partial amount of their eggs at different times throughout the spawning season. Darters use three egg-laying strategies that include egg burying, egg attaching, and egg clustering. Egg burying is accomplished when the female partially buries her body in the substrate and the male mounts her and fertilizes the eggs as they are expelled. Egg attaching requires the eggs to be attached to a fixed object in the water such as sticks, rocks, or plant roots. In this case, the male follows the female and when a suitable object is located, he mounts the female and fertilizes the eggs as they are attached to the selected object a few at a time. Egg clustering involves the female depositing the eggs in a shelter. The male protects the eggs by clearing debris and silt. The eggs are fertilized as they are deposited in the shelter. The male then guards the shelter until the eggs mature. Due to lack of observations it is currently unknown which reproduction strategy *E. sellare* uses.

1.5 The Maryland Darter

Etheostoma sellare (Radcilffe & Welsh, 1913)
Maryland darter

Hadropterus sellaris: Radcliffe and Welsh, 1913:29-32 (original description, type locality, Swan Creek).

Poecilichthys sellaris: Hubbs & Black, 1940:3 (comparison with *P. variatus group)*

Etheostoma (Etheostoma) sellare: Bailey & Gosline, 1955:16, 39(vertebrae).- Knapp et al., 1963:455(rediscovery). -Collette, 1965:588 (nontuberculate).- Richards, 1966:823-827 (relationships).-Collette & Knapp, 1966:48(types)

Etheostoma sellare is a member of genus *Etheostoma*, subgenus *Etheostoma* in the family Percidae.

1.5.1 Systematic Position

Radcliffe and Welsh (1913) placed the Maryland darter in the genus *Hadropterus* without comment. The lack of caduceus scales led Hubbs and Black (1940) to reassign *E. sellare* to the genus *Poecilichthys*. Bailey and Gosline (1955) placed *E. sellare* in the subgenus *Etheostoma* along with 13 other species. Richards (1966) separated the subgenus into three species groups (*E. blennioides, E. thalassinum, and E. variatum*) and two specialized relatives *E. blennius and E. sellare*. The evidence of a specialized triangular head shape and a naked belly led Tsai (1966) to state "*E. sellare* diverged early from other members in the evolution of the subgenus *Etheostoma.*" Knapp (1976) agreed with Richards (1966) in that *E. sellare* does not fit well in the subgenus *Etheostoma* and went as far to say that it could be placed in the genus *Percina* (except for the lack of caduceus scales) or possibly placed in a monotypic subgenus containing only *E. sellare*. A lack of further anatomical, osteological, or electrophoretic characteristics have prevented this movement from occurring. Most recently, Burr (1979) placed *E. sellare* in a group (within subgenus *Etheostoma)* with *E. inscriptum* and *E. thalassinum* based on 51 morphological characters (Figure 1). Burr (1979) noted that there are obvious discrepancies with the placement of *E. sellare* but considering current research, nothing has justified a movement from this group.

1.5.2 Description

Etheostoma sellare is distinguishable from other darters by having asymmetrical caudal fins and a large head. This species has a maximum length of 70 mm and ctenoid scales. Knapp et. al. (1976) described the distinguishing characteristics as follows: lateral line is straight and complete with 43-53 scales; infraorbital and supratemporal canals complete; preoperculomandibular pores 10; brachiostegal membranes slightly conjoined; each with six brachiostegals; snout moderately produced; premaxillary frenum present; pelvic fins long, separated by a space equal to ¾ or more of each fin base; anus surrounded by blunt striated lobes, not finger-like villi; preopercular margin entire; anal fin with two spines, 2^{nd} relatively weak; palatine and vomerine teeth well developed. The presence of four dark, anteriorly directed dorsal saddles is the most obvious identifying characteristic of the species.

1.5.3 Diet

Unlike many other darter species, the diet of *E. sellare* is, at least seasonally, largely comprised of snails. A study conducted in 1976 by Knapp examined the diet of 35 individuals. It is shown in this study that relatively large numbers of snails were consumed. Of the 35 specimens examined, 28 had ingested from 1 to 11 snails. This study concluded that the diet of the

Maryland darter is largely comprised of caddis flies (*Hydropsyche*) and snails (probably *Somatogyrus virginica*) at least seasonally. Very little information exists on the habits and ranges of members of the Hydrobiidae (mud snails) family. According to Knapp (1976) *S. virginica* occupies vegetation and rocks in silty substrates in rivers and the mouths of tributaries. The only other darter taxa known to consume snails in quantity are the members of the *Percina* subgenus *Imostoma*. The broad head and wide gape of *E. sellare* may allow for ingestion of snails.

1.5.4 Reproduction

Little is known about the Maryland darter's reproductive behaviors. No breeding specimens have been collected. Spawning occurs in late April or early May, according to the reproductive status of collected specimens (Knapp, 1976). Females develop a swollen white pad on the urogenital papilla, while the male papilla is short and shows little seasonal variation.

1.5.5 Habitat

The Maryland darter is known only from three locations (Deer Creek, Gashey's Run and Swan Creek) in Harford County Maryland (Figure 2). The Swan Creek location is vague and no specimens have been observed in Swan Creek since its type-collection in 1912. The Deer Creek location appears to be the first substantial riffle upstream from the mouth of the stream. The Gashey's Run location near the Oakington Road bridge seems to be somewhat farther upstream than the lowest riffle of that stream. However both confirmed locations (Deer Creek and Gashey's Run) occur on or near the "Fall line" (Figure 3) as these streams flow from the Piedmont to the Coastal Plain physiographic Province. At the Deer Creek location the riffle is large (25m wide) while the Gashey's Run location is much smaller (approximately 5m wide). Both locations share a dense canopy of bank-side trees that shade the stream.

1.5.6 History

The Maryland darter was described in 1913 from two juvenile specimens taken from Swan Creek near Havre de Grace, Maryland. The collection and description of *E. sellare* (originally *Hadropterus sellaris*) was made by naturalists Lewis Radcliffe and William W. Welsh. The researchers described the type locality as, "*water 6 inches deep, on a long, stony riffle, where the bottom was comparatively free from bowlders and the current so swift that one would not have expected to find fishes of any kind.*" After its description, sampling efforts increased in an attempt to obtain more specimens of *E. sellare*. However, all surveying efforts failed for over 50 years until 1962 when a group of Cornell University graduate students discovered a juvenile Maryland dater among a group of tessellated darters taken from nearby Gashey's Run. The species then evaded researchers again until 1965 when a single female was found in Gashey's Run by the same group of researchers (Knapp et al., 1963). Later that same year *E. sellare* was found nearly 15 river miles away from the Swan Creek/Gashey's Run area. Raney and Schwartz found the species in Deer Creek near the intersection of Stafford Road and Craig's Corner Road approximately 1.1 miles upstream from the mainstem of the Susquehanna River (Knapp, 1976). During this effort 33 Maryland darters were collected in May of 1965. Further searching in this area that same year yielded an additional 38 specimens (Knapp, 1976). This location appeared to support the largest population of *E. sellare* ever recorded. Historical reports suggest that this is the last known location to have supported an *E. sellare* population (Raesly 1991). At the time, it was apparent that Deer Creek represented the Maryland darter's optimal habitat. In recognition of its rarity, the Maryland darter is a federally endangered species (U.S. Fish and Wildlife Service, 1967).

In 1974, the Environmental Protection Agency (EPA) issued a permit, despite considerable opposition, for the release of chlorinated wastes into a headwater stream of Deer Creek, called Ebaugh's Creek near Stewartstown, Pennsylvania, upstream from the historic Deer Creek location. Unfortunately, no stream integrity data exists for Deer Creek prior to 1974, making it impossible to accurately quantify the effect this discharge has had on the last known location of *E. sellare.* Critical habitat was established for the Maryland darter on August 29, 1984 (U.S. Fish and Wildlife Service, 1984) (Figure 4). In 1985, the United States Fish and Wildlife Service (USFWS) published the recovery plan for the Maryland darter. The plan states that six separate stable populations of *E. sellare* must be located before, downlisting the species to threatened (U.S. Fish and Wildlife Service, 1985). Between 1986 and 1988 Richard Raesly observed 8 Maryland darters at the Deer Creek (Stafford Bridge location) (Raesly pers. comm.).

In 1990 the Maryland Natural Heritage program funded a survey to obtain information on the Maryland darter in the Deer Creek watershed. They selected 10 sites along the lower 35 miles of Deer Creek. This survey located no Maryland darters. During 1991 and 1992, Raesly conducted surveys restricted to the riffle just below the Stafford Bridge on Deer Creek. These surveys captured 5,579 fish representing 35 species, but no Maryland darters were observed (Raesly, 1991; Raesly, 1992). In July 1995, the USFWS received a petition to delist the Maryland darter from the Maryland Farm Bureau, Inc., of Randallstown, Maryland. Section 4(b)(3)(A) of the Endangered Species Act of 1973 requires that the USFWS make a finding on whether a petition to list, delist, or reclassify a species presents substantial scientific or commercial information to demonstrate that the petitioned action may be warranted. The USFWS determined that the petition did not present sufficient data for the Maryland darter to be ruled extinct. The USFWS cited Etnier (1994) stating that “it is not uncommon for large time periods to pass between observations of rare species”. Within this statement the USFWS stated that “While the failure to find the Maryland darter in Deer Creek provides evidence that the species has declined in Deer Creek and may be extirpated (at least temporarily) there, it does not provide sufficient evidence to declare the species extinct. The species may continue to survive in the Susquehanna River adjacent to Deer Creek. To date, this area has not been extensively searched because of the very difficult sampling conditions there. Until this area has been adequately searched, we cannot rule out the survival of the Maryland darter there (Federal Register / Vol. 61, No. 32 / Thursday, February 15, 1996)." In 2007 the USFWS published a 5-year review of the Maryland darter. Since the last observation was in 1988 the USFWS determined no new data had been presented on the species.

Maryland Department of Natural Resources and the U.S. Fish and Wildlife Service have implemented a recovery plan (U.S. Fish and Wildlife Service, 1985) with goals of managing streamside buffer zones to reduce sedimentation and agricultural pollution along with maintenance and improvement of water quality. However, a lack of available resources has slowed the recovery plan implementation process. In 2008 USFWS began funding research for sampling at all the historical locations and in the Susquehanna mainstem. Maryland Department of Natural Resources, Marshall University, and Frostburg State University conducted surveys in all these locations.

Large sections of the Susquehanna mainstem remain to be searched near the Conowingo Dam (Harford County, Maryland). The extreme flow fluctuations produced by Conowingo Dam make this habitat difficult to sample effectively. Survey work in the mainstem should be a top priority.

Recent efforts to delist the darter are ill-founded without additional survey work (Neeley et al., 2003).

1.6 Sampling Rare Species

Portions of all ecosystems are made of rare species that have small local populations (McArdle, 1990). These species pose problems for researchers studying diversity and local populations. Studies have attempted to quantify the probability of occurrence for rare species (Levins & Culver, 1971, McArdle, 1990, Bayley & Peterson, 2001). Sampling for rare species presents unique problems for researchers. The probability of capturing a rare species is low for any given sampling event. For mobile species like fish, the probability of capture is even lower. Determining when a rare species is not present in an area is only possible with a 100% exhaustive survey, (e.g. a census) which is impractical. There is a probabilistic relationship between sampling units/effort, rarity of the species, and exhaustiveness of the survey (McArdle, 1990). Estimates for species richness or presence should be representative of the organisms that are there. However, observation or capture-based surveys often underestimate rare species (Bayley & Peterson, 2001), but it is logical that as the number of sampling units increase the probability of detecting a rare species also increases (McArdle, 1990).

Many species have been found years after they were ruled extinct. The Carolina Parakeet, Bermuda Petrel, La Palma Giant Lizard, Horton Plains Slender Loris, Cave Splayfoot Salamander, Red-Limbed Mount Nimba Reed

Frog, Omaniundu Reed Frog, Kunimasu trout, harlequin darter, saddleback darter, striped back darter, and gilt darter are all examples of species that were once thought to be extinct, later to be rediscovered.

1.7 Project Significance

The main objectives of this study were to (1) search for a live specimen of *E. sellare* and (2) determine if habitat similar to the last known location still exists.

This study provides information on the status of *Etheostoma sellare.* This species has been proposed for delisting. The USFWS has determined that insufficient data exists for the ruling of extinction for *E. sellare*. Since *E. sellare* is a rare species sampling for specimens is difficult and the probability of capturing one on any given sampling event is incredibly low. This study attempts to shed light on the reasons behind the lack of information for this species and provide analysis using current techniques and technology that was not previously available.

2.0 Methods

Fishes were surveyed at four tributary sites using backpack electro-fishing and visual surveys. Large sections of the Susquehanna River were surveyed for fish using a combination of visual surveys and benthic trawling. The main purpose of all fish surveys was to collect a live specimen of *Etheostoma sellare*. Sampling took place over four sampling periods between August 2008 and July 2010. During the searching efforts habitat data were collected in an effort to develop a collection of parameters that would indicate suitable habitat for *E. sellare*. Historical reports and Maryland Biological Stream Survey Data (1994-2009) were used in analysis of suitability for all sites when available.

2.1 Site Selection

Sampling sites were selected from historical reports (Radcilffe & Welsh, 1913; Knapp, 1976) of known *E. sellare* locations. In an effort to sample a variety of habitat types, sampling in the mainstem was targeted. (Susquehanna mainstem-**Error! Reference source not found.**, Deer Creek-Figure 66, Swan Creek-Figure 77, Gashey's Run-Figure 88, Octoraro Creek-Figure 99).

2.2 Electro-fishing

Surveys of fishes were conducted at all tributary sites (Deer Creek, Swan Creek, Gashey's Run, and Octoraro Creek) using a backpack electro-fisher (Smith-Root Model 15A, operating at a frequency of 60 Hz; variable voltage). Collections were made using a 1.5m x 3.05m, 0.3mm mesh seine as a block net positioned 3m to 6m downstream from the backpack fisher. Field crews ranged in size from three to ten people to increase the efficiency of substrate disturbance in the sample area. The backpack fisher and assistants moved downstream disturbing the substrate to dislodge benthic fishes and corral fish into the block net positioned downstream. Each tributary site was sampled completely in congruent parallel transects during each of the three sampling periods. Each fish collected was identified, counted, and then released. Counts of each species per seine haul were recorded for species abundance analysis.

2.3 Trawling

Trawls were conducted on a timed basis ranging from 1 to 2 minutes. If the trawl became obstructed or snagged during the sampling period, time was stopped until the net was freed and trawling was able to continue. A Self-Contained Underwater Breathing Apparatus (SCUBA) diver freed snagged trawls that were too severe to be retrieved from the boat.

Trawls used in this study included modified Missouri-type 2.44 m wide trawl and a modified Missouri-type 2.44m wide electric trawl (Figure 10). The specifics for both nets are as follows: the nets had a cod end 2.14m in length with 1.5mm diameter nylon twine. The bar mesh was 19.05mm and lined with a 3.18mm mesh. The body consisted of No. 7 (8.87 cm^2) high-density sapphire polyethylene netting and the bag was made of No. 12 (3.175 cm^2) high-density polyethylene netting (Herzog et al. 2005). The trawl was connected to "otter" boards or "planer" boards, which were 40.64 cm by 34.12 cm pieces of weighed wood. The "otter" boards helped keep the trawl net open horizontally, while a float line and sink line kept the trawl opened vertically. The trawl boards were equipped with steel runners on the bottom to help with drag and durability.

Trawls were deployed from the bow of the boat while the boat was facing upstream. After the net was in the water the boat slowly traveled in reverse for the allotted sampling period. The boat speed was just faster than the current to ensure the opening of the trawl and to maintain contact with the bottom of the river. Samplers dragged the trawl across the bottom, and the sink line and "otter" boards dislodge rocks and debris exposing small-bodied benthic fish. The trawl was designed to expose benthic fishes, such as darters, from their hiding places.

A modified electro-fishing research vessel towed the trawl. The trawl was attached to two hard points on the bow of the boat with 9.5mm polydacron line that was 20 meters in length. The length of the rope used during trawling varied by the depth of the site and the speed of the boat. On average an effective trawl consisted of a 2 to 1 rope to depth ratio. A marker buoy was attached to the end of the codpiece with a 2m braided nylon rope. This was done to provide a visual marker of where the trawl was at all times. In addition to serving as a marker for the trawl, the tail buoy also helped in the retrieval of snagged trawls.

Latitude and longitude coordinates, temperature, and average depth were recorded using a Garmin 480 model C GPS at the start and end of each trawl. The duration, distance from the shore, and rope length were also recorded on data sheets.

Fish collected from each trawl were identified then immediately released. Data from every trawl were placed into a database at the Maryland Department of Natural Resources and Marshall University.

2.4 Snail Abundance Surveys

A one square meter Surber frame was used to determine the boundaries for snail collections. Samplers positioned the metal Surber frame on the bottom of the stream and collection sites were spaced at 150m to 200m intervals from the first substantial riffle to the mouth of each sampled tributary. Divers equipped with mask and snorkel collected all visible snails within the one square meter frame. Divers placed collected snails in 70% ethanol solution for preservation. Snails were taken to Marshall University's Aquatic Laboratory for enumeration and identification to species.

2.5 Habitat

Habitat data were collected within the same reaches as electro-fishing and snail sampling (Figure 11-Deer Creek, Figure 12-Octoraro Creek, Figure 13-Swan Creek, Figure 14-Gashey's Run). Sampling began at the farthest upstream location and moved downstream in a sinuous pattern to the end of the reach. Three types of habitat metrics were measured during this project. A Marsh McBirney FLO-MATE portable flowmeter was used to record velocities to the nearest tenth of a meter per second and depth. Substrate types were classified according to six categories: fines, sand, gravel, cobble, boulder, and bedrock. Dominant substrate was determined visually at the same location of the velocity sample.

2.6 Visual Surveys

2.6.1 Mainstem

The area below the Conowingo Dam is highly variable habitat. During certain levels of regulated release large areas of the mainstem are inaccessible by boat. Visual surveys were conducted in an effort to assess these areas for possible habitat capable of supporting an *E. sellare* population. Visual surveys were conducted in the mainstem of the Susquehanna River below the Conowingo dam. Visual Surveys were conducted over two days in July 2010 (7/8/2010 & 7/9/2010). The area surveyed consisted of an approximately 50 m wide reach on the right descending bank going from the fisherman's access below the Conowingo

Dam to the confluence of Rock Run and the Susquehanna River (5.2kilometers). The survey area was divided up into approximately 75m sub-reaches and the data for each sub-reach was recorded at the end of the reach. Five divers were spaced approximately 5m apart and moved downstream in approximately 100m intervals parallel to the bank. At the end of each interval coordinates, fish abundance score, algae coverage score, snail abundance score, mussel abundance score, relative velocity, average depth, and dominant substrate type were recorded (Figure 15).

2.6.2 Tributaries

Visual surveys were also conducted at the Stafford Bridge crossing of Deer Creek (Figure 16), the mouth of Deer Creek (Figure 17), and below the Moore road railroad bridge on Octoraro Creek to the mouth (Figure 18). Surveying at these locations were less structured and divers covered as much area as was needed for complete coverage of the site. Divers moved up stream in roughly parallel transects. Divers then made a complete species list of observed species.

2.6.3 Data Analysis

2.6.3.1 Statistical Analysis

All abiotic factors recorded at the stream sites were averaged to get mean values for each metric at each site.

Principle Components Analysis (PCA) was performed on the electro-fishing catch data to show any correlations between the site's fish communities. This analysis attempts to reduce the degree of duplication or correlation in a dataset by finding highly correlated combinations of factors. Each combination is called a component. These components are represented in the two axes of the PCA output. Any sites grouped closely together are similar, whereas sites far apart are relatively dissimilar. Factors are represented by lines in the PCA ordination. The line length is a measure of importance of the value. The line direction also can be used to draw conclusions based on where sites are relative to the line

The abundance of each species captured at each electro-fishing site was calculated for each sampling event. Using PC-ORD 5 software a PCA was run to compare the site's fish communities. The species of fishes were used as the factors and each site was used as a record. The sampling events were analyzed separately and combined to show trends or relationships between the sites based on the makeup of their fish community.

The number of snails at each collection location was calculated to provide an average number of snails per square meter for each sampled stream. The average number of snails per square meter was used to compare the sample streams.

Statistical analysis for the comparison of the two types of trawling was conducted in Microsoft Excel 2007 using the "TTEST" function. This allowed us to show if one method of trawling was more efficient and which of the two types of trawl used (electrified or non-electrified) was statistically better at capturing fish. Since it was undetermined which data set (electrified or non-electrified) had unequal variance a 2-tailed t-test was used. A P-value of 0.05 or less was considered significant.

2.6.3.2 GIS Analysis

For each area of interest images were created with ArcMap 9.3 (ESRI 2009) using Inverse Distance Weighting (IDW) for each recorded characteristic. IDW is a technique that interpolates a surface based on known points. Any unknown points on the raster are calculated using values from the 16 nearest neighbor values. A polygon was created for each area of interest using a best-fit polygon that encompassed the points. The mask was used to clip each IDW raster file to show only those interpolated values within the sample reach.

This process was used to create maps showing the distribution of values for each metric recorded at each visual survey location and for distribution of fishes captured in the mainstem.

Land use classification was executed for each watershed in which stream electro-fishing was conducted. Land cover types were separated into five categories: Open water, developed, barren land, forest, and agriculture. The categories were developed using information from the National Land Cover Database (NLCD) maintained by the Multi-Resolution Land Characteristics Consortium (MRLCC). Open water included all areas with generally less than 25% cover of vegetation or soil and areas that are periodically saturated or covered with water. Developed land was categorized by a range of areas with a mixture of constructed materials and vegetation (low intensity) up to areas that include areas where people reside or work in high numbers (high intensity). Barren land included areas of bedrock, desert pavement, strip mines, gravel pits and other accumulations of earthen material. Agriculture included areas of grasses, legumes, or grass-legume mixtures planted for livestock grazing or the production of seed or hay crops, typically on a perennial cycle and areas used for the production of annual crops, such as corn, soybeans, vegetables, tobacco, and cotton, and also perennial woody crops such as orchards and vineyards. Polygons were created to use as a mask for each watershed. Land cover data was mined from the NLCD which is maintained by the MRLCC. A raster file was obtained from the National Seamless Data Server and reclassified in ARC 9.3 for analysis. Once the raster file was reclassified an extraction was executed for each watershed using the boundary polygon as a mask. Pixel counts for each classified land use type were obtained from the attribute tables and entered into Excel for further analysis. A table showing the land use percentages for each watershed by land use classification is shown in Table 18.

Data was obtained from the Maryland Department of Natural Resources (MDDNR) Maryland Biological Stream Survey (MBSS) for two counties in Maryland (Harford and Cecil). Data provided biological integrity index scores for fish and benthic macroinvertebrates. The locations of stream surveys were projected in Arcmap 9.3 and IDWs were created based on index scores (Figure 52, Figure 53, Figure 54, Figure 55, Figure 56, and Figure 57). County boundaries were used for a mask to extract the resulting IDW maps. The maps were then analyzed using the minus function, spatial analyst tool, to show change in fish index scores between each round (Figure 58, Figure 59, and Figure 60).

3.0 Results

3.1 Stream Electro-Fishing

Length of stream reach sampled varied with each site based on suitable habitat and accessibility (Table 1). The electro-fishing survey yielded forty-one species of fish (10,488 individuals) which were collected in the electro-fishing portion of this study. A summary of the numbers of fishes captured by species, location, and sampling date is shown in Table 22. More fish were captured at the Swan Creek and Gashey's Run sites in November 2009 than the Deer Creek and Octoraro Creek Sites. This was reversed in the summer of 2010 with more fish being captured at the Deer Creek and Octoraro Creek sties. A summary of seine hauls at each site on each date are shown in Table 22. At the Deer Creek and Octoraro Creek locations the numbers of fish per seine haul increased from November 2009 to the June and July 2010 sampling periods. Again, this trend was reversed for the Swan Creek and Gashey's Run locations. The summary of fishes per seine haul by sampling date is shown in Table 3. The numbers of total fishes captured at each site varied (426 to 2262 individuals) between each sampling date and location. No Maryland darters were captured during this study. Principal Component Analysis (PCA) was run on the electro-fishing data to show the relationship between the sites based on the composition of their fish community. As previously stated the fish community data was time sensitive so in an effort to maintain validity each sampling period was viewed as a separate data set. The November 2009 PCA showed strong similarities between the Deer Creek and Octoraro Creek sites (percent cumulative variance = 90.681) (Figure 19). The June 2010 data did not show the strong relationship between Deer and Octoraro Creek (percent cumulative variance = 82.711) (Figure 20). Instead the fish community in Octoraro Creek was more similar to that of Swan Creek in June 2010. PCA was determined inappropriate for the July 2010 data since only Deer and Octoraro Creeks were surveyed.

3.1.1 Darters

Five species (*Etheostoma blennioides, E. olmstedi, E. zonale, Percina bimaculata,* and *P. pelta*) of darters (2,250 individuals) were captured during the electro-fishing portion of this study. Over the 3 sampling periods the most darters were captured at the Deer Creek site (969 individuals) followed by Octoraro Creek (844 individuals), Swan Creek (403 individuals), and Gashey's Run (34 individuals). A summary of the darters captured at each site by date is shown in Table 4. The percentage of darters that made up each fish community varied from 0.15% at Gashey's Run November 2009 (lowest) to 43.66% at Deer Creek July 2010 (highest). The percentage of darters in each fish community is shown in Table 5.

3.1.1.1 Deer Creek

The Deer Creek site below the Stafford Road Bridge was sampled on three occasions: November 7th 2009, June 3rd 2010, and July 8th 2010. This site proved to be the most diverse with 31 species (4179 individuals) being captured. This site showed the most variation of abundance (November 2009- 426 individuals to June 2010- 2262 individuals). *Anguilla rostrata* was the most abundant species (2025 individuals) followed by *Etheostoma olmstedi* (820 individuals). This site had the greatest number and diversity of darters with all five species represented in the 969 individuals. At Deer Creek 23.19% of the fish captured were darters. The summary of fishes captured by species and date is shown in Table 22.

Velocity, depth, and substrate values were measured on June 4th and July 7th 2010. The results of these measurements are shown Table 6 and Table 7.

3.1.1.2 Octoraro Creek

The Octoraro Creek site above the Susquehanna River Road Bridge was sampled on three occasions: November 7th 2009, June 6th 2010, and July 7th 2010. Sampling produced 29 species of fish (2501 individuals). *Anguilla rostrata* was the most abundant species at the site (1144 individuals) followed by *E. olmstedi* (747 individuals) being captured over the three sampling events. At this site all five species of darter were captured

(844 individuals). Octoraro Creek had the highest percentage of darters captured during the three sampling events at 33.73%. Table 2 provides a summary of fishes captured by species and date.

Velocity, depth, and substrate values were measured on June 4th and July 7th 2010 (Tables 8 and 9).

3.1.1.3 Swan Creek

The Swan Creek site was sampled on two occasions: November 8th 2009 and June 3rd 2010. Sampling produced 22 species of fish (2033 individuals). *Semotilus atromaculatus* was the most abundant species (456 individuals) followed by *E. olmstedi* (402 individuals) at the site. This site had fewer darter species than the Deer Creek and Octoraro Creek sites with only 403 individuals representing two species, 402 were *E. olmstedi.* One *P. bimaculata* was captured in November 2009 at this site. Of the fish captured at Swan Creek 19.82% were darters. Table 2 shows the summary of fishes captured by species and date.

Velocity, depth, and substrate values were measured on June 4th 2010. The results of these measurements are shown in Table 10.

3.1.1.4 Gashey's Run

The Gashey's Run site was sampled on two occasions: November 8th 2009 and June 3rd 2010. This site showed the lowest level of diversity and abundance with 18 species of fish (1774 individuals). *Fundulus diaphanus* was the most abundant species (827 individuals), followed by *Lepomis gibbosus* (420 individuals). All 827 *F. diaphanus* were captured in November 2009. This site also showed the lowest diversity of darters with *E. olmstedi* (34 individual) being the only species of darter captured at this site. Gashey's Run had the lowest percentage of darters in the fish community (1.92%). Table 2 shows the summary of fishes captured by species and date.

Velocity, depth, and substrate values were measured on June 4th. The results of the measurements is shown in Table 11.

3.2 Trawling

Benthic trawling was conducted one week each summer for three years (2008, 2009, and 2010). Electrified trawling was conducted exclusively in 2008. On two occasions (2009 and 2010) both electrified and non-electrified trawls were used to cover as much area in a variety of habitats as possible. Table 12 shows the summary of the data collected with both electrified and non-electrified trawls. Overall 153 trawls were conducted during this study. The 103 electrified trawls produced 22 fish species. The 50 non-electrified trawls produced 13 fish species. *Percina bimaculata, A. rostrata, L. auritus, M. dolomieu, I. punctatus, L. cyanellus, M. americana, N. insignis, P. marinus,* and *L. xanthurus* were captured in electrified trawls and not in non-electrified trawls. *Pimephales notatus* and *Ambloplites rupestris* were captured in non-electrified trawls and they were not captured in electrified trawls. Electrified trawls averaged 412.9 fish per working hour.

Non-electrified trawls averaged 121.0 fish per hour. Electrified trawls averaged 307.3 darters per working hour and non-electrified trawls averaged 70.1 darters per working hour. Electrified trawls captured eight benthic fish species (E*. olmstedi, P. bimacualta, E. zonale, P. peltata, A. nebulosus, I. punctatus*, *N. insignis,* and *P. marinus*). Non-electrified trawls captured only four benthic species (*E. olmstedi, E. zonale, P. peltata, and A. nebulosus*). During the approximately 17,407 working seconds that trawls (electrified and non-electrified) were deployed 1572 fish were captured representing 24 species; 4 of which were darter species (*E. olmstedi, E. zonale, P.bimaculata, P. peltata).*

Electrified trawling was significantly better at capturing darters species (P-value = 0.0003), benthic fish species (P-value = 0.005), and total fish (P-value = 0.0074) when compared to the non-electrified trawl.

3.3 Visual Surveys

Visual surveys totaled 307.1 man-hours of searching during this survey and included large sections of the mainstem, Deer Creek, and Octoraro Creeks were surveyed.

3.3.1 Deer Creek and Octoraro Creek

Visual surveys in Deer Creek yielded two additional species (*Morone saxatilis* and *Sander vitreus*) not previously observed in electro-fishing. Octoraro Creek visual surveys produced one species (*Cyprinus carpio*) that was not observed during electro-fishing.

3.3.2 Mainstem

The visual surveys resulted in 136 sub-reaches being assessed. Figure 21 shows a map of all visual survey. ArcMap software was used to create maps showing the distribution of values for each metric recorded during the visual surveys. Visual survey area substrate map is shown in

Figure 22. Bathometric Map of the visual survey area is shown in FFigure 23. Distribution of fish abundances are shown in Figure 24. Distributions of snail abundances are shown in Figure 25. Distribution of algae abundance is shown in Figure 26. Distribution of mussel abundance is shown in Figure 27.

3.4 Snail Abundance Surveys

Field crews conducted snail abundance surveys over two days in July (July 8th 2010 and July 9th 2010). During these surveys 664 individuals were collected from 21 locations in two streams (Deer Creek and Octoraro Creek). Three species were identified from the collected snails. *Gonibasis* (*Elimia*) *virginica* was the most abundant (89.6% of all snails collected). *Gonibasis* (*E.*) *virginica* specimens produced both smooth and lirate shell morphs. *Leptoxis carinata* was the second most abundant snail collected. Five specimens of a small Hydrobidae species (probably *Amnicola limosa*) were collected. All three species were found in Deer Creek. Octoraro Creek yielded only two species (*Leptoxis carinata and* Hydrobidae species (probably *Amnicola limosa*)). In Deer Creek 483 snails were collected over 13 sites while in Octoraro Creek 181 snails were collected over nine locations. Deer Creek sites averaged 37.15 snails per square meter and Octoraro Creek averaged 20.11 snails per square meter. The Inverse Distance Weighting (IDW) tool in the Arcmap software was used to create snail distribution maps for the surveyed areas (Figure 44 and Figure 45). Table 13 shows a summary of snails collected at each location.

3.5 Land use

Land use classification was executed for each watershed in which stream electro-fishing was conducted. A table showing the land use percentages for each watershed by land use classification is shown in Table 18. Agriculture made up the largest percentage of total land cover in all the sampled watersheds. Forest land cover accounted for the second largest portion of all the sampled watersheds.

The Deer Creek watershed consisted of 0.83% open water/wetland, 2.54% developed land, 0.58% barren land, 33.43% forest, and 62.62% agriculture (Figure 47).

The Octoraro Creek watershed consisted of 2.55% open water/wetland, 2.95% developed land, 0.91% barren land, 20.56% forest, and 73.03% agriculture (Figure 48).

The Swan Creek watershed consisted of 9.56% open water/wetland, 15.07% developed land, 0.49% barren land, 34.13% forest, and 40.76% agriculture (Figure 49).

The Gashey's Run watershed consisted of 10.79% open water/wetland, 12.71% developed land, 0.49% barren land, 28.30% forest, and 47.70% agriculture (Figure 50).

3.6 Historical Data

Using MBSS, data maps were created showing distribution of fish and benthic macroinvertebrate index scores for two selected counties in Maryland. Maps were also created showing change in fish scores over time. The maps showing change between each round show areas in the two counties that seem to be improving and other areas that are in decline. In particular the Deer Creek area seems to be improving from rounds 1 to 3 while Octoraro creek, Swan Creek, and Gashey's Run seem to be in decline (Figure 60). Other areas along the mainstem of the Susquehanna appear to be improving as well.

4.0 Discussion

4.1 Stream Electro-fishing

4.1.1 Deer Creek

The vast majority (>90%) of collected *E. sellare* specimens were found in Deer Creek. The Deer Creek site (below the Stafford Bridge) is the last known (1988) location where *E. sellare* was observed (Raesly, 1991). Thus this site is the last site known to have habitat capable of supporting a population of *E. sellare.* Since this site has not changed physically since the darter's last observance, it is considered a reference site for typical *E. sellare* habitat (Raesly pers. comm.). If other locations have habitat similar to the Deer Creek site, then it is not unreasonable to predict that these sites are also capable of supporting a population of *E. sellare.*

Little is known about *E. sellare's* movements or reproductive habits. Other darter species of the same genus (*Etheostoma*) range in movements from local movements throughout the lifetime of a darter to migrations of several miles for spawning (Reed, 1968; Scalet, 1973; Winn, 1958). Scalet (1973) studied the movements of the Orange Belly darter (*Etheostoma radiosum*) and found that this particular species is thought to have a small (<100m^2) home range. Reed 1968 commented on the movement of eight darter species in Northwestern Pennsylvania streams and found that a low recapture rate of darters was a result of low seining efficiency or darter migration. Conversely, Winn (1958) notes that some darters (*E. spectabile, E. blennioides, E. nigrum, and H. maculates)* migrated several miles from their permanent habitat during spawning events. Winn (1958) also commented on the disappearance of darters from riffles after the breeding season. This phenomenon of darter migration is possibly the reason for the apparent loss of *E. sellare* from the Deer Creek riffle. It is possible that the Deer Creek riffle represents a temporary breeding ground for the Maryland darter. If the biotic integrity of this site has been compromised then it is possible that the site no longer meets the requirements for *E. sellare* reproduction thus justifying the reason that the darter has not been captured there for many years.

Between September 1990 and December 1992, Raesly sampled the Deer Creek site on 12 separate dates. The most noticeable difference between past and current surveys is the number of *A. rostrata* captured. The number of *A. rostrata* greatly increased from the early 90s (314 individuals) to the recent surveys (2025 individuals). This is probably due to the migratory behavior of the American Eel (A. rostrata) and the different sampling methods used in the surveys. The 2009 and 2010 surveys were backpack electro-fishing surveys while the majority of the 1990-1992 surveys did not use the electro-fisher.

Aside from the number of American Eels captured there is little difference between the fish collected at the Deer Creek riffle in the early 90s to the recent surveys. A summary of all the fishes collected in each respective survey is shown in Table 14. If eels are taken from both data sets the values of percent darters, percent tolerant species, percent invasive species, percent generalists omnivores invertivores, and percent lithophillic spawners are within a few percent of each other (Table 15). A table showing the comparison of the two surveys when eels are excluded from the analysis is shown in Table 16. Although the comparison appears to show, close similarities between the two surveys, it must be considered that the data from the earlier surveys were collected over 12 sampling events while the most recent data were collected on just 3 sampling events. The higher catch per unit effort for the most recent surveys is attributed to the use of the backpack electro-fishing unit. In most recent surveys a much larger abundance of fishes were captured; however, the makeup of the fish community here appears to have changed little in the 16 years between the surveys.

4.1.2 Octoraro Creek

This site was surveyed due to its proximity and similar habitat attributes to the Deer Creek site. Octoraro Creek is the most similar of the sites to the Deer Creek site (Figure 33, Figure 34, Figure 35). The PCA's of the stream fish communities grouped Octoraro and Deer Creek very closely in November 2009 (Figure 19) but this relationship was not present in the summer 2010 (Figure 20). The similar fish communities in the winter are thought to be a result of a period of low fish movement that occurs in the winter months. This loss of similar fish communities is most likely due to the availability of the sites to migrating fishes. The Octoraro Creek site is much closer to the out flow of the Conowingo Dam that may be making it difficult for fishes to move into this area. The summer 2010 PCA groups Octoraro Creek loosely with Swan Creek, a site that is also somewhat isolated from fish immigration due to the extensive deposits of fine sediment at its mouth (Figure 28). Although no specimens of *E. sellare* have been collected or observed from this site its similarities to Deer Creek means that it may be a site that *E. sellare* could utilize.

4.1.3 Gashey's Run

Aside from the Deer Creek location Gashey's Run is the only other confirmed location for the Maryland darter. Fifty-one years after its description *E. sellare* evaded researchers until it was discovered there by researchers from Cornell University in 1962 (Knapp,1963). However Gashey's Run has been impacted more than any other *E. sellare* location in recent years (Raesly pers. comm.). This impact is most likely due to the large amount of urbanization occurring within the watershed boundaries (Figure 29). Evidence of erosion and sediment deposition are obvious when surveying this location (Figure 30 and Figure 31). During summer low flows Gashey's Run consists of only a few small pools connected by tiny channels sometimes only 30 cm in width (Figure 32). This site is only accessible by new fish populations via the Chesapeake Bay which is subject to tidal fluctuations and migration from here to the Oakington Bridge (a known historic location of *E. sellare)* would be unlikely due to the large area of degraded habitat that would have to be traversed. Even if the unlikely immigration of new populations occurred the Gashey's Run site now represents heavily impacted habitat and it is unlikely to support a population of *E. sellare*. The large amounts of erosion and sedimentation occurring here are most likely responsible for the loss of this site as a potential *E. sellare* location.

4.1.4 Swan Creek

Swan Creek is the type locality for *E. sellare.* However the exact location is unknown. Radcilffe and Welsh (1913) described the location as follows *"the species described within were seined in Swan Creek, near Havre de Grace, Md., in water 6 inches deep, on a long, stony riffle, where the bottom was comparatively free from bowlders and the current so swift that one would not have expected to find fishes of any kind."* Since the description no additional specimens have been collected in Swan Creek proper. However three specimens have been collected in nearby Gashey's Run (a tributary to Swan Creek). The mean substrate value in Swan Creek proper is similar to that of the Deer Creek reference site; however it lacks the diversity (Figure 33). The lack of observations at this site are possibly due to the large amounts of sediment deposition occurring at the mouth of the watershed. If the type locality was within the lower 2.5 kilometers then sediment deposits have likely covered it. Relatively low densities of darters and other intolerant species may be due to the isolation of the watershed due to the large flats (approximately 5 kilometers in length) of silt deposits that species would have to traverse to reach the site. The presence of intolerant migratory species (*Dorosoma cepedianum*) supports this hypothesis. Further research is needed to determine the reasons for the lack of darters in this community.

4.2 Trawling

The effectiveness of benthic trawling can vary based on a number of parameters such as substrate, depth of water, in-river obstructions, boat speed, and reaction of fish to the trawl (Sheehan & Rasmussen, 1999). When trawling in depths of less than 1.5m the effectiveness of trawling was noticeably lower. This is thought to be due to “prop scare” or scaring fish from the area due to turbulence from the boat motor as the boat travels in reverse, preventing a representative sample from being taken. The area below the Conowingo dam consists of very diverse habitat. Boulders, submerged logs, and undercut bedrock are common in this area. This abundance of submerged structure makes trawling efficiently difficult. However samplers did not count snagged or partially obstructed trawls. During this study the electrified trawl was much more efficient at capturing fishes than the non-electrified trawl. The electrified trawl captured 3.4 times more fish per working hour than the non-electrified trawl. The electrified trawl also detected twice as many benthic species as the non-electrified trawl. The use of an electrified trawl when possible for future *E. sellare* searching efforts is recommended.

Utilizing the data collected from complete trawls fish and darter abundance maps were created in Arcmap (Figure 44 and Figure 45). These maps show areas that seem to have higher relative densities of fishes and darters. The fish "hotspots" appear to be located in proximity to the mouths of tributaries (Deer Creek and Rock Run). This distribution can most likely be attributed to the diversity of habitat associated with the junction of streams. In this study, Deer Creek and Rock Run confluences had the highest fish densities. Trawling in similar areas to the "hotspots" found in this study is likely to yield similar results.

Some researchers believe that *E. sellare* may be a large river darter because the only known populations were found in the lowest riffle of tributary streams. However the mainstem of the Susquehanna River below Conowingo Dam is a very dynamic area and difficult to sample. Benthic trawling is essentially the only way to survey fish communities in some of this area. Benthic trawling (especially electrified trawling) should be continued in the mainstem in future surveying efforts. Considering that the electrified trawl was significantly more efficient at capturing fish in all categories, it is logical to recommend that an electrified trawl be used whenever possible. However, it needs to be considered that the lower sample size of non-electrified trawls is likely responsible for the dramatic significance between the two trawling types. However this relationship will most likely stand with further research.

4.3 Visual Surveys

The mainstem of the Susquehanna below the Conowingo Dam represents a highly variable habitat. Since the dam controls this area this section can experience rapid changes in water level, flow, and velocity. During certain periods of these fluctuations habitat similar to that of the Deer Creek reference site occurs. Further research is needed to quantify the amount of habitat that is created or lost during different release rates from the dam. In particular one section surveyed during this study may contain potential *E. sellare* habitat. The area near the right descending bank approximately 3.0 kilometers below the fisherman's access and 2.0 kilometers upstream from the junction of Deer Creek and the mainstem may represent habitat suitable for *E. sellare* (Figure 38). This area had higher diversity and abundances of all the biotic parameters (fish abundance, snail abundance, mussel abundance) measured during the survey. Whether or not fish continue to utilize this habitat during higher release events is unknown. It appears that areas behind large boulders could provide refuge for species incapable of withstanding the force of the higher release rates from the dam. These areas contained finer substrate than areas exposed to the full force of the released water. These areas show similar diversity of substrate to the Deer Creek and Octoraro Creek sites.

4.4 Snail Abundance Survey

Snails were collected from Deer and Octoraro Creeks during this study. Deer Creek had a higher mean number of snails per m^2. The snail density data was used to create IDW maps for both sampled streams (Deer Creek-Figure 44, Octoraro Creek-Figure 45). There are noticeable areas on both sampled streams that show a rapid increase in snail density. The area in Deer Creek where snail density increases is approximately 0.8 kilometers upstream from the mouth. This roughly corresponds with the area where water is pushed back up into the stream during releases from the Conowingo dam. However, directly below this area was a noticeable change in substrate from cobble and unconsolidated gravel to sand. In Octoraro Creek the area where snail density makes this change is approximately 0.5 kilometers from the mouth. During a snorkel survey, water backed up well above the change in snail density. It is unknown whether the snails are attempting to move downstream and are being stopped by the warmer water, change in substrate, or some other reasons that is causing the snails to congregate here.

4.5 Additional Analysis

4.5.1 Fish Stranding

Due to rapid decreases in flow between power production events approximately 380,556m^2 of riverbed becomes dewatered below the dam (Figure 39, Table 17). An area of particular interest is the mouth of Octoraro

Creek due to its proximity to the outflow of the Conowingo Dam (Figure 40). Since such a large area of formally underwater habitat becomes exposed, there is potential for fish stranding on gravel bars or being trapped in side-channels or pools.

Evidence of fish stranding was observed on July 6th and 9th 2010. On July 6th and 9th 2010 researchers searched portions of the area below the Conowingo Dam for evidence of stranded fishes. In one small pool (approximately 2m^2) five *E. olmstedi* and one *P. bimaculata* were found deceased. Researchers believe that the cause of death was due to the increased temperature (34.4 °C) of the small isolated pool. On that same day researchers also found six shad (*Clupeidae* sp.) dead on an exposed shoal approximately 100m from the previously noted pool. Figure 41 shows the locations of these dead fishes. Further research is needed to quantify standing potential and the numbers of fish likely stranded by each change in flow.

4.5.2 Flow Reversal in Streams

During the visual surveys at the mouth of Deer Creek there was an observed temporary flow reversal in lower Deer Creek on July 7, 2010 (starting at 5:15pm). It was later discovered that this flow reversal was the effect of a release from the Conowingo Dam. During the initial stages of this particular release the flow of the lower 1.07 kilometers of Deer Creek was actually reversed or stopped and water moved in an upstream direction at velocities reaching 0.65 cubic feet per second (cfs). Further research is needed to determine the effects of this water being pushed upstream. It is possible that this flow reversal would result in increased deposition in the lower reaches of Deer Creek. The temperature of the mainstem near the mouth of Deer Creek was considerably warmer than the water flowing from Deer Creek. It is also possible that warmer water from the mainstem being pushed upstream could affect the biota of the lower reaches. No measurements of flow reversal were taken in Octoraro Creek, however flow reversal was observed on July 6th 2010.

Since this phenomenon is occurring in Deer Creek (5.5 kilometers) and Octoraro Creek (1.5 kilometers) which are relatively near the Conowingo Dam, it is likely occurring in the lower reaches of other tributaries below the dam. As previously stated, further research is needed to determine the effects of this flow reversal on stream biota and the local habitat.

4.5.3 Land Use

The type of land use occurring in a watershed has been shown to have direct effects on the stream biota (Allan, 2004; Allan et al., 1997; Fausch et al., 2002; Huston, 2005; Inwood, 2002; Schlosser, 1991; Towsend et al., 2003). By utilizing the data collected from the land cover maps I was able to make conclusions on the potential health of a watershed (King et al., 2005).

The lower Susquehanna was one of the first areas in America to be settled by Europeans, so it not surprising that such a large percentage of the sampled watersheds is agriculture. However, it is interesting that such a large portion of land-cover remains in agricultural use when these watersheds are in such proximity to very large human populations (Baltimore). As large farms are being divided and sold either for agriculture use or for development, the land becomes more intensively used (Burcher & Benfield, 2006). This can lead to faster rates of erosion and increased sediment deposition in streams. Studies have shown that agriculture can have major effects on stream biota including causing increased nutrient input, sediment deposition, increased velocities, and higher amounts of pollution (del Rosario et al., 2002; Dodds et al., 2004; Neill et al., 2001). Fortunately, privately owned farms provide a certain amount of protection from development of these watersheds. However, the large percentage of agriculture is most likely responsible for a majority of the sedimentation occurring in the lower Susquehanna River basin.

The relatively low percentages of developed land for the sampled watershed are likely an underestimate. Deer and Octoraro Creeks are much larger watersheds (442,373 kilometers^2 and 413,436 kilometers^2 respectively) than Swan Creek and Gashey's Run (43,714 kilometers^2 and 11,302 kilometers^2 respectively). The higher percentage of developed land in Swan Creek and Gashey's Run watersheds is likely a reflection of the small size and proximity to major highways. Although, studies have shown that when as little as 1% of a watershed's total area becomes impervious, that some low tolerance species disappear from that drainage (Stranko, 2010). Since the surveyed area is among the fastest growing in the nation it is likely that amounts of developed land, and impervious surface, will increase rapidly in the coming years.

4.5.4 Historical Data

The maps showing change between each round show areas in the two counties that seem to be improving in stream integrity and other areas that are in decline. In particular, the lower Deer Creek watershed seems to be improving from rounds 1 to 3 while Octoraro creek, Swan Creek, and Gashey's Run seem to be in decline (Figure 60). Other areas along the mainstem of the Susquehanna appear to be improving as well. These locations would be candidate areas of locations to look for populations of *E. sellare.* Since so little is known about the Maryland darter's habitat requirements the exact amount of impact the species can tolerate is unknown. Therefore areas that show no change or improving fish IBI scores, and are similar to the Deer Creek location, are possible areas that are capable of supporting *E. sellare* populations.

5.0 Conclusions

The main objectives of this study were to (1) search for a live specimen of *E. sellare* and (2) determine if habitat similar to the last known location still exists.

Overall results from the study yielded no Maryland darter specimens. The reason for this is most likely due to a combination of factors. Low probability of occurrence and the highly variable habitat as a result of rapid changes in flow regime are compounding factors in the effort to locate a specimen of this species. Because this study was only conducted over four weeks throughout three years the absence of the Maryland darter from this study is relatively insignificant. A negative result is not conclusive since it is not unusual for long periods to pass between observations of a rare species (Ethier, 1994).

Etheostoma sellare is a species that needs to be considered carefully because species with small populations are vulnerable and at an increased risk of extinction (Primack, 2006). How anthropogenic impacts are affecting this species is vital to understanding the reasons behind its sporadic appearance in Maryland's streams. Previous impacts (i.e. pollution discharges) may have had impacts on this species in the past. Because of its limited range, seemingly small impacts could have large repercussions on this species population. The possibility of competition from other closely related darter species (*E. omlstedi, E. zonale, and E. blennioides)* may also be contributing to the apparent absence of this species.

Areas similar to the Deer Creek site were found in the mainstem. Therefore it is not unreasonable to consider that the species may be using these areas instead of the former locations where the species was found. Knowledge of the natural history of *E. sellare* is very limited. The movements and spawning behavior of this species are essentially unknown. The areas where *E. sellare* has been previously located may represent temporary or marginal habitat used by the species. If those sites biological integrity are now compromised then it is possible that the species is no longer capable of surviving at those locations.

Continual degradation of the lower Susquehanna watershed could result in extirpation of the species if that has not already occurred. Further searching in areas with similar habitat to that of the Deer Creek site is needed. Based on results from this study, further research is required before the species is considered extinct.

5.1 Future Research Recommendations

Future research efforts should focus on the historical sites and areas in the mainstem with similar habitat attributes of the Deer Creek site observed in this study. Electrified trawling proved to be the most efficient and reasonable way of sampling the mainstem, this method should be implemented in future searching efforts. In order to focus on areas of similar habitat attributes to Deer Creek a habitat mapping in the mainstem would provide a rapid assessment of large portions of the substrate of the mainstem and then sampling could be focused in the areas that share similar depth and substrate characteristics with Deer Creek.

Two phenomenona that were observed during this study deserve additional attention. The effects of back flow in the lower sections of tributaries below the dam may be isolating these streams from new populations of stream biota, by forming a warm water or substrate barrier. Also the possibility for fish to become stranded is high in the area near the mouth of Octoraro Creek. Additional efforts to quantify the numbers and types of fishes that are at risk of becoming stranded is needed to assess the implication of this phenomena occurring.

LITERATURE CITED

Allan, J .D., Erickson, D.L. , & Fay, J. (1997). The influence of catchment land use on stream integrity across multiple spatial scales. *Freshwater Biology (37),*149-61.

Allan, D.J. (2004). Landscapes and Riverscapes: The Influence of Land Use on Stream Ecosystems. *Annual Review of Ecology, Evolution, and Systematics, (35)* 257-284.

Bailey, R.M., & Gosline, W.A.. (1955) Variation and systematic signifigance of vertevral counts in the American fishes of the family Percidae. Museumn of Zoology, *University of Michigan Miscellaneous Publications, (93)*, 44.

Boward, E., Kazyak, P., Stranko, S., Hurd, M., & Prochask, A. (1999). From the mountains to the sea: The state of Maryland's freshwater streams. United States Environmental Protection Agency, Atlantic Ecology Division, Narragnsett, Rhode Island and Maryland Department of Natural Resources, Annapolis, Maryland, USA.

Bradford, M. J. (1997) An experimental study of stranding of juvenile salmonids on gravel bars and in sidechannels during rapid flow decreases. *Regulated Rivers: Research & Management, (13),* 395–401.

Burcher C.L. & Benfield, E.F. (2006) Physical and Biological Responses of Streams to Suborbanization of Historically Agricultural Watersheds. *Journal of the North American Benthological Society, (25)2,* 356-369.

Burr, M. B. (1979) Systematics and Life History Aspects of the Percid Fish *Etheostoma blennius* with Description of a New Subspecies from the Seqatchie River, Tennessee. *Copeia, (2*), 191-203.

Collette, B.B. (1965) Systematic signifigance of breeding tubercles in fishes of the family Percidae. *Proceedings of the U.S. Natural History Museum, 117 (3518),* 567-614.

Collette, B.B., & Knapp, L.W. (1966) Catalog of the type specimens of the darters (Pisces, Percidae, Etheostomatini). *Proceeding of the Natural History Museum, 119(3550),* 1-88.

del Rosario, R.B., Betts, E.A., Resh, V.H. (2002) Cow Maunre in Headwater Streams: Tracing Aquatic Insect Responses to Organic Enrichment. *Journal of the North American Benthological Society, (21)2,* 278-289.

Dodds W.K., Gido, K., Whilles, M.R., Fritz, K.M., Matthews, W.J. (2004) Life On the Edge: The ecology of Great Plains Prairie Streams. BioScience. (54)3: 205-216.

Dynesis, M., Nilsson, C. (1994) Fragmentations and flow regulation of river systems in the northern third of the world. *Science, (266),* 753–762.

Etnier, D. A. (1994) Our southeastern fishes- what we have lost and what we are likely to lose. *Proceedings of the Southeastern Fishes Council, (29),* 5-9.

Fausch K. D., Torgersen, C. E., Baxter, C. V., Li, H. W. (2002) Landscapes to riverscapes: bridging the gap between research and conservation of stream fishes. *BioScience, (52),* 483- 98

García, A., Jorde, K., Habit, E., Caamaño, D., & Parra, O. (2009) Downstream environmental effects of dam operations: Changes in habitat quality for native fish species. *River Research and Applications.*

Helfman, G.S., Collette, B.B. & Facey, D.E. (1997). The Diversity of Fishes. Blackwell Science, Malden, Massachusetts.

Hern, A. J. (2006) “The Current Status and Future Prospect of Selected Rare Fish Species in Maryland.” M. S. Thesis. Univresity, Frostburg, Maryland. 52-66.

Herzog, D.P., Barko, V.A., Scheibe, J.S., Hrabik, R.A., & Ostendorf D.E. (2005). Effiency of a Benthic Trawl for Sampling Small Bodied Fishes in Large River Systems. *North American Journal of Fisheries Management, (25),* 594-603

Hocutt, C.H. & Wiley, E.O. editors (1986). The Zoogeography of North American Fishes. John Wiley and Sons, Inc. New York.

Hubbs, C.L. & Black J.D. (1940) Percid fishes realated to *Poecilichthys variatus,* with descriptions of three new forms. *Occasional Papers to the University of Michigan Museum of Zoology, (416),* 30.

Huston, M. A. (2005) The Three Phases of Land-Use Change: Implications for Biodiversity. *Ecological Applications, (15)6,* 1864-1878.

Inwood, S.E., Tank, J.L., Bernot, M.J. (2005) Patterns of Denitrification Associated with Land Use in 9 Midwestern Headwaster Streams. *Journal of the North American Benthological Society, (24)2,* 227-245.

Irvine, R. L., T. Oussoren, J.S., Baxter, & Schmidt, D.C. (2009) The effects of flow reduction rates on fish stranding in British Columbia, Canada. *River Research and Applications, (25),* 405–415.

King R.S., Baker, M.E., Whigman, D.F., Weller, D.E., Jordan, T.E., Kazyak, P.F., Hurd M.K. (2005) Spatial Considerations for Linking Watershed Land Cover to Ecological Indicators in Streams. *Ecological Applications, (15)1,* 137-153.

Knapp, L.W. (1976) Redescription, relationships and status of the Maryland darter, *Etheostoma sellare* (Radcliffe and Welsh), an endangered species. *Proceeding of the Biological Society of Washington, (89),* 99-118.

Knapp, L. W., Richards, W.L., Miller, R.V., & Foster N.R. (1963) Rediscovery of the percid fish *Etheostoma sellare* (Radcliffe and Welch) *Copeia*, 455.

Kuehne, R.A. & Barbour R.W. (1983) *The American Darters*. Lexington, KY: The University Press of Kentucky, Lexington KY.

Layman, S. R., Simons, A.M., & Wood, R.M. (1993) Status survey of the Dirty Darter (*Etheostoma olivaceum*) and the Bluemask Darter

(*Etheostoma* (*Doration*) sp.) with notes on the fishes of the Caney Fork River system, Tennessee. *Journal of Tennessee Academy of Science, (68*), 65–70.

Levins, R. & Culver, D. 1971. Regional Coexistence of Species and Competition between Rare Species. *Proceedings of the National Academy of Science, 68(6)*, 1246-1248.

Mattingly, H.T. & Galat, D.L. (2002) Distributional Patters of the Threatened Niangua Darter, *Etheostoma nianguae,* at Three Spatial Scales, with Implications for Species Conservation *Copeia, (3),* 573-585.

McArdle, B.H. (1990) "When Are Rare Species Not There." *Oikos, 57(2),* 276-277.

McKinney, M.L. (2001) Effects of human population, area, and time on non -native plant and fish diversity in the United States. *Biological Conservation, (100),* 243–252.

McKnight, J. (2011) Maryland Department of Natural Resources. Wildlife and Heritage Service. *Invasive and Exotic Species*. Maryland Department of

Natural Resources. Web.5Jan2011 <http://dnr.maryland.gov/wildlife/plants_wildlife/invintro.asp>.

Miller, R.R., Williams, J.D. & Williams, J.E. (1989) Extinctions of North American fishes during the past century. *Fisheries (Bethesda*), *14(6),* 22-38.

Morita, K. & Yamamoto, S. (2002) Effects of Habitat Fragmentation by Damming on the Persistence of Stream-Dwelling Charr Populations. *Conservation Biology, (16),* 1318–1323.

Neeley, D. A., Hunter, A.E., & Mayden, R.L. (2003) Threatened Fishes of the World: *Etheostoma Sellare* (Radcliffe & Welsh) 1913 (Percidae). *Environmental Biology of Fishes, (67),* 340.

Neill C., Deegan L.A., Thomas S.M., & Cerri C.C. (2001) Deforestation for Pasture Alters Nitrogen and Phosphorus in Small Amazonain Streams. *Ecological Applications, 6(11),* 1817-1828.

Page, L.M. (1983) Handbook of Darters. TFH Publishing, Neptue City, NJ. 271.

Primack, R.B. (2006) Essentials of Conservation Biology. Fourth Edition.

Sinauer Associates, Inc. 188-196.

Pringle, C. M., Freeman, M.C., & Freeman, B.J. (2000) Regional effects of hydrologic alterations on riverine macrobiota in the New World: tropical-temperate comparisons. *BioScience, (50),* 807–823.

Radcliffe, L & Welsh, W.W. (1913) Description of a new darter from Maryland. *Bulliten U.S. Bureau of Fish, (32),* 29-32.

Raesly, R.L. (1991) Population Status of the Endangered Maryland Darter, *Etheostoma sellare,* in Deer Creek. Report submitted to the Maryland Natural Heritage Program 28.

Raesly, R.L. (1992) Population Status of the Endangered Maryland Darter, *Etheostoma sellare,* in Deer Creek. Report submitted to the Maryland Natural Heritage Program 13.

Reed, R. T. (1968) Mark and Recapture Studies of Eight Species of Darters (Pisces: Percidae) in Three Streams of Northwestern Pennsylvania. *Copeia,(1),* 172-175.

Richards, W.J. (1966) Systematics of the percid fishes of the *Etheostoma* *thalassium* species group with comments on the sub genus *Etheostoma. Copeia, (4),* 823-838.

Roth, N.E., Southerland, M.T., Mercurio, G., Chaillou, J.C., Kayzak, P.F., Stranko, S., Prochaska, A.P., Heimbuch, D.G., Seibel J.C. (1999) State of the Streams: 1995-1997 Maryland Biolgical Stream Survey Results. Maryland Department of Natural Resources, Annapolis, Maryland, USA.

Saltveit, S., Halleraker, J., Arnekleiv, J., and Harby, A. (20010 Field experiments on stranding in juvenile atlantic salmon (*Salmo salar*) and brown trout (*Salmo trutta*) during rapid flow decreases caused by hydropeaking. *Regulated Rivers: Research & Management, (17),* 609–622.

Saunders, D.A. & Hobbs, R.J. (1991) Biological consequences of ecosystem fragmentation: a review. *Conservation Biology, (5),* 18–32.

Scalet, C.G. (1973) Stream Movements and population Density of the Orangebelly Darter, *Etheostoma radiosum cyanorum* (Osteicthyes: Percidae*). The Southwestern Naturalist, 17(4),* 381-387.

Schlosser, I.J. (1991) Stream fish ecology: a land-scape perspective. *BioScience, (41),* 704-12.

Sheehan, R.J., and J. Rasmussen. (1999) Large Rivers *in* C. C. Kohler and W. A. Hubert, editors. *Inland Fisheries Management in North America, 2nd edition. American Fisheries Society,* 529-559.

Stranahan, S.Q. (19930 *Susquehanna, River of Dreams*. Baltimore: Johns Hopkins University Press.

Stranko, S.A. & Klauda, R. J. (2010) "Maryland's Streams, Imperiled by Urbanization." Maryland Department of Natural Resources, Monitoring and Non-Tidal Assessment Division, Annapolis, Maryland, USA.

Tiemann, J.S., Gillette, D.P., Wildhaberr, M.L., & Edds, E.D. (2004) Effects of lowhead ddams on riffle-dwelling fishes and macroinvertibrates in a Midwestern River. *Transactions of the American Fisheries Society, (133),* 705-717.

Townsend C.R., Doledec S., Norris R., Peacock K., Arbuckle C. (2003) The influence of scale and geography on relationships between stream community composition and landscape vari-ables: description and prediction. *Freshwater Biology, (48),* 768-85.

Travnichek, V.H., Bain, M.B., & Maceina, M.J. (1995) “Recovery of a Warmwater Fish Assemblage after the Initiation of a Minimum-Flow Release Downstream from a Hydroelectric Dam.” *Transactions of the American Fisheries Society, (124),* 836-844.

Tsai, Chu-Fa, (1966) *in* Knapp (1976) A study of the systematic of Ehteostoma zonale (Cope) and its relatives, and the phylogeny of subgenus Etheostoma Rafinesque (Percidae, Teleostei), Ph.D. thesis, Cornell Universtiy, Ithaca NY 365.

U.S. Fish and Wildlife Service. (1984) Designation of critical Habitat for the endangered Maryland darter. *Federal Register, (49),* 34228-34232.

U.S. Fish and Wildlife, Interior. (1996) "Endangered and Threatened Wildlife." *Federal Register, (61.32),* 5971.

U.S Fish and Wildlife Service. (1967) Native fish and wildlife; endangered species. *Federal Register, (32),* 4001.

U.S. Fish and Wildlife Service. (1985) The Maryland darter recovery plan. USFWS, Newton Corner MA, 39.

Wilcox, B. A., & Murphy, D. (1985) Conservation strategy: the effect of fragmentation on extinction. *The American Naturalist, (125),* 879–887.

Wine, M.S., Weston, M.R., & Johnson, R.L. (2008) Density Dynamics of a Threatened Species of Darter at Spatial and Temporal Scales. *Southeastern Naturalist, 7(4),* 665-678.

Winn, H.E. (1958) Comparative Reproductive Behavior and Ecology of Fourteen Species of Darters (Pisces-Percidae). *Ecology Monographs, 28(2),* 155-191.

Winston, M.R., Taylor, C.M., Pigg, J. (1991) Upstream extirpations of four minnow species due to the damming of a prairie stream. *Transactions of the American Fisheries Society, (120),* 98-105.

Appendix A

Tables

Site	Sample Reach Length	Mean Width
Deer Creek	115m	30m
Octoraro Creek	150m	40m
Gashey's Run	350m	7m
Swan Creek	180m	10m

Table 1: Electro-fishing reach length and width sorted by site

SITE	DC1109	DC610	DC710	OC1109	OC610	OC710	GR1109	GR610	SC1109	SC610	
Seine Hauls	112	231	161	104	55	79	59	36	56	64	
Species											Totals
A. rostrata	113	1389	523	109	452	583	2	69	55	104	3399
E. olmstedi	81	144	595	126	32	589	2	32	327	75	2003
C. spiloptera	0	49	80	6	15	54	0	0	17	13	234
C. commersonii	0	19	45	0	20	68	0	73	1	37	263
E. zonale	13	18	7	22	6	21	0	0	0	0	87
N. insignis	61	210	8	40	27	16	0	0	9	33	404
R. atratulus	3	29	5	0	0	9	21	76	0	357	500
R. cataractae	67	155	66	33	0	51	0	0	0	0	372
E. maxillingua	0	11	9	0	0	2	0	0	63	114	199
N. micropogon	46	90	66	33	13	12	0	0	1	0	261
N. analostanus	0	5	2	0	0	0	0	0	3	15	25
N. hudsonius	0	15	1	12	2	2	0	1	0	0	33
F. diaphanus	6	1	0	18	0	0	827	0	152	0	1004
L. cornutus	0	6	0	2	0	0	0	1	14	11	34
P. bimaculata	3	36	20	2	1	5	0	0	1	0	68
C. caeruleomentum	13	10	0	0	0	0	0	0	0	0	23
L. auritus	0	7	3	0	2	3	1	5	0	4	25
N. procne	2	1	2	33	2	6	39	14	32	6	137
P. peltata	14	9	28	14	4	21	0	0	0	0	90
N. rubellus	0	3	4	0	0	1	0	0	0	0	8
C. funduloides	0	16	0	0	0	0	0	0	0	14	30
N. nebulosus	0	1	0	0	0	0	2	44	1	13	61
S. trutta	0	1	0	0	0	0	0	0	0	0	1
P. marinus	0	1	0	0	0	0	0	0	0	0	1
L. cyanellus	0	0	0	0	0	2	0	19	0	5	26
L. gibbosus	1	0	0	1	2	2	378	42	81	11	518
S. atromaculatus	0	0	0	0	0	0	1	8	451	5	465
E. oblongus	0	0	0	0	0	0	1	39	0	1	41
L. macrochirus	0	0	0	3	0	1	67	3	1	0	75

N. crysoleucas	0	0	0	0	0	0	0	1	0	0	1
H. nigricans	3	0	7	1	0	5	0	0	5	1	22
M. americana	0	0	0	0	6	0	0	0	0	0	6
M. salmoides	0	0	11	0	0	4	3	0	0	0	18
D. cepedianum	0	0	0	0	0	0	3	0	0	0	3
A. natalis	0	0	0	0	0	1	0	0	0	0	1
M. dolomieu	0	0	5	0	0	1	0	0	0	0	6
M. saxatilis	0	0	0	0	0	1	0	0	0	0	1
I. puntatus	0	0	0	0	0	1	0	0	0	0	1
A. rupestris	0	0	0	0	0	1	0	0	0	0	1
E. blennoides	0	0	1	0	0	1	0	0	0	0	2
S. corporalis	0	0	3	0	0	0	0	0	0	0	3

Table 2: Summary of fishes captured sorted by site and date of capture.

Site	Fish/Seine Haul
GS1109	22.8305
GR610	11.8611
SC1109	21.6786
SC610	12.7969
OC1109	4.3750
OC610	10.6182
OC710	18.5190
DC1109	3.8036
DC610	9.6364
DC710	9.2609

Table 3: Summary of fish captured per seine haul sorted by site and date of capture.

Darters	E. olmstedi	E. zonale	P. bimaculata	P. peltata	E. blennoides	Total Darters
Site						
DC1109	81	13	3	14	0	111
DC610	144	18	36	9	0	207
DC710	595	7	20	28	1	651
OC1109	126	22	2	14	0	164
OC610	32	6	1	4	0	43
OC710	589	21	5	21	1	637
GR1109	2	0	0	0	0	2
GR610	32	0	0	0	0	32
SC1109	327	0	1	0	0	328
SC610	75	0	0	0	0	75

Table 4: Summary of Darters captured sorted by species and site

Site	Date	Darters	Total Fish Captured	% Darters
GS1109	Nov_09	2	1347	0.0015
GR610	Jun-10	32	427	0.0749
SC1109	Nov-09	328	1214	0.2702
SC610	Jun-10	75	819	0.0916
OC1109	Nov-09	164	455	0.3604
OC610	Jun-10	43	584	0.0736
OC710	Jul-10	637	1463	0.4354
DC1109	Nov-09	111	426	0.2606
DC610	Jun-10	207	2262	0.0915
DC710	Jul-10	651	1491	0.4366

Table 5: Summary of darter percentage of each site’s fish community sorted by site and date

Descriptive Statistics

	N	Minimum	Maximum	Mean	Std. Deviation
Velocity (cfs)	34	.07	4.00	1.6038	1.02398
Depth (decimal ft.)	34	1.00	9.80	4.4412	2.03352
Substrate	34	2.00	5.00	3.7059	.75996
Valid N (listwise)	34				

Table 6: Summary of Deer Creek abiotic factors measured in June 2010

Descriptive Statistics

	N	Minimum	Maximum	Mean	Std. Deviation
Velocity (cfs)	33	.20	4.30	1.3473	.98458
Depth (decimal ft.)	33	1.00	6.50	3.0515	1.30219
Substrate	33	2.00	5.00	3.7576	.66287
Valid N (listwise)	33				

Table 7: Summary of Deer Creek abiotic factors measured in July 2010

Descriptive Statistics

	N	Minimum	Maximum	Mean	Std. Deviation
Velocity (cfs)	21	.35	2.50	1.4648	.70769
Depth (decimal ft.)	21	.40	7.20	3.1333	1.86610
Substrate	21	2.00	6.00	3.1905	.74960
Valid N (listwise)	21				

Table 8: Summary of Octoraro Creek abiotic factors measured in June 2010

Descriptive Statistics

	N	Minimum	Maximum	Mean	Std. Deviation
Velocity (cfs)	17	-.03	2.65	.9171	.63431
Depth (decimal ft.)	17	1.30	4.10	2.8824	.77559
Substrate	17	1.00	4.00	3.0588	1.24853
Valid N (listwise)	17				

Table 9: Summary of Octoraro Creek abiotic factors measured in July 2010

Descriptive Statistics

	N	Minimum	Maximum	Mean	Std. Deviation
Velocity (cfs)	14	.04	2.55	.9671	.77968
Depth (decimal ft.)	14	1.00	3.80	1.9000	.85844
Substrate	14	3.00	4.00	3.8571	.36314
Valid N (listwise)	14				

Table 10: Summary of Swan Creek abiotic factors measured in June 2010

Descriptive Statistics

	N	Minimum	Maximum	Mean	Std. Deviation
Velocity (cfs)	28	.01	2.58	.5832	.70373
Depth (decimal ft)	28	.20	8.20	2.3071	2.04558
Substrate score	28	2.00	4.00	2.8929	.62889
Valid N (listwise)	28				

Table 11: Summary of Gashey's Run abiotic factors measured in June 2010

	Electrified-trawl	Non-Electrified-trawl	Combined
Trawls	103	50	153
Duration (sec)	12172	5235	17407
Avg Duration (sec)	118.2	104.7	115.9
Total Species	22	13	24
Total Catch	1396	176	1572
Darter Species	4	3	4
Benthic Fish Species	8	4	8
Fish per Working Hour	412.9	121.0	337.0
Darters Per Working Hour	307.3	70.1	239.6

Table 12: Summary of Electric vs. Non-electric Trawls

Stream	Waypoint	*Gonibasis* (*Elimia*) *virginica*	*Leptoxis carinata*	Hydrobidae species
Octoraro Creek	271	14		
Octoraro Creek	272	32		
Octoraro Creek	276	23		
Octoraro Creek	278	40		
Octoraro Creek	277	43		
Octoraro Creek	279	15		
Octoraro Creek	284	1		
Octoraro Creek	285	12		1
Deer Creek	251	1		
Deer Creek	254	1		
Deer Creek	258	3		
Deer Creek	259	35	9	
Deer Creek	260	39	12	1
Deer Creek	261	14	4	
Deer Creek	262	8	7	
Deer Creek	263	192	27	3
Deer Creek	264	48	1	
Deer Creek	265	6	1	
Deer Creek	266	16	3	
Deer Creek	267	17		
Deer Creek	268	35		

Table 13: Summary of snails collected sorted by site

	1990-1992	2009-2010
Petromyzon marinus	14	1
Anguilla rostrata	314	2025
Alosa mediocris	1	0
Clinostomus funduloides	21	16
Cyprinella analostana	61	7
Cyprinella spiloptera	130	129
Cyprinus carpio	6	0
Exoglossum maxillingua	21	20
Luxilus cornutus	15	6
Nocomis micropogon	249	202
Notropis analostanus	61	7
Notropis hudsonius	32	16
Notropis rubellus	32	7
Notropis procne	2	5
Pimephales notatus	11	0
Rhinichthys atratulus	69	37
Rhinichthys cataractae	76	288
Semotilus atromaculatus	3	0
Semotilus corporalis	3	3
Catostomus commersoni	21	64
Hypentelium nigricans	80	10
Moxostoma spp.	1	0
Fundulus diaphanus	0	7
Ameiurus nebulosus	0	1
Noturus insignis	40	279
Ambloplites rupestris	2	0
Lepomis auritus	21	10
Lepomis cyanellus	15	0
Lepomis gibbosus	68	1
Lepomis macrochirus	34	0
Micropterus dolomieui	4	5
Micropterus salmoides	0	11
Etheostoma olmstedi	979	820
Etheostoma zonale	46	38
Etheostoma blennioides	0	1
Perca flavescens	2	0
Percina bimaculata	20	59

Percina peltata	21	51
Cottus caeruleomentum	45	23
Salmo trutta	0	1

Table 14: Comparison between fish captured by Rasely (early 90's) surveys and recent surveys from this study

	1990-1992	2009-2010
Total Fish	2520	4150
Total Darters	1066	969
% Darters	0.4230	0.2335
% Tolerant Species	0.476190476	0.225060241
% invasive species	0.042460317	0.013253012
% Gen, OM, IV	0.903174603	0.895180723
% Lithophilic spawners	0.237301587	0.124096386

Table 15: Summary of calculated Fish metrics between Rasely (early 90s) surveys and recent surveys from this study

	1990-1992	2009-2010
Total Fish	2206	2125
Total Darters	1066	969
% Darters	0.4832	0.4560
% Tolerant Species	0.543970988	0.439529412
% invasive species	0.04850408	0.025882353
% Lithophilic spawners	0.271078876	0.242352941

Table 16: Summary of calculated fish metrics between Rasely (early 90s) surveys and recent surveys from this study without American Eels

Total Mainstem Area Below Dam	18,838.284
Habitat at high flow	17,637.167km²
Habitat at low flow	17,256.610km²
Area of Potential Habitat Lost	380.556km²
% of total area lost	0.0214067

Table 17: Summary of change in exposed riverbed between high and low flow events.

Land Use Type	Octoraro Creek	Deer Creek	Gashey's Run	Swan Creek
Open Water/Wetlands	2.55%	0.83%	10.79%	9.56%
Developed	2.95%	2.54%	12.71%	15.07%
Barren Land	0.91%	0.58%	0.49%	0.49%
Forest	20.56%	33.43%	28.30%	34.13%
Agriculture	73.03%	62.62%	47.70%	40.76%

Table 18: Table of land cover percentages separated by watershed and land cover type.

Appendix B
Figures

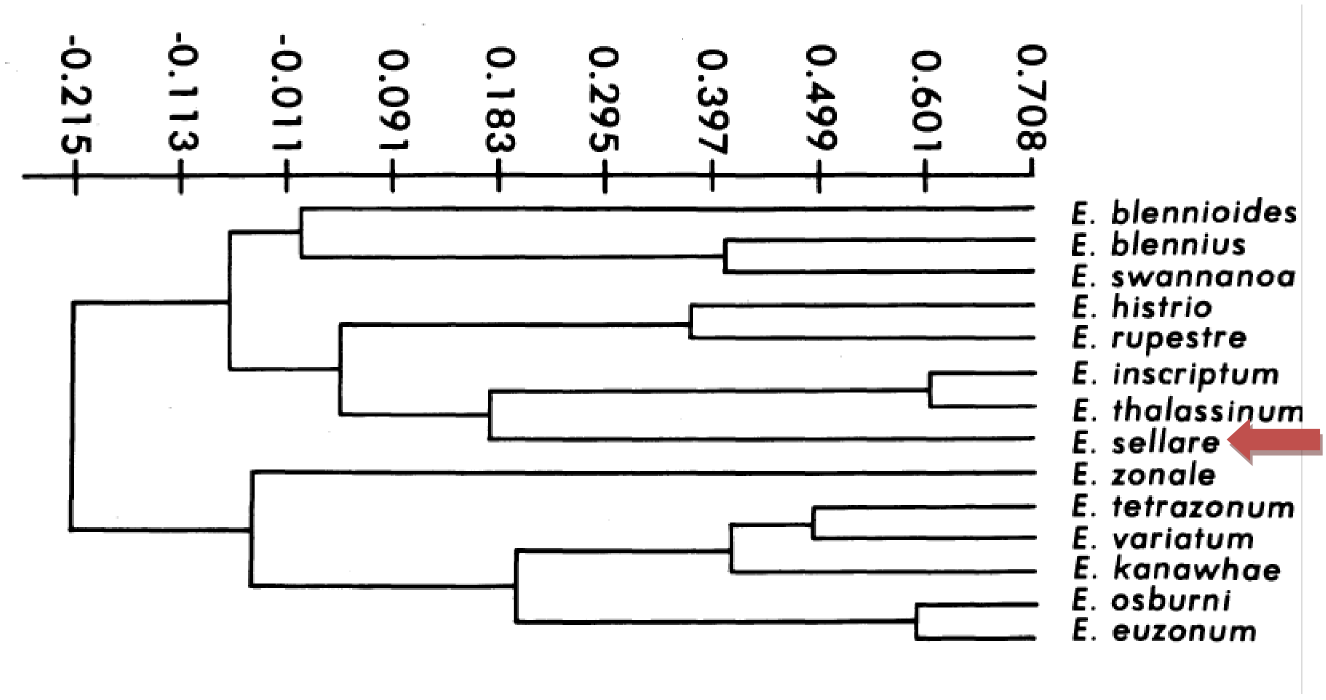


Figure 1: Systematic position of *E. sellare* according to Burr (1979).

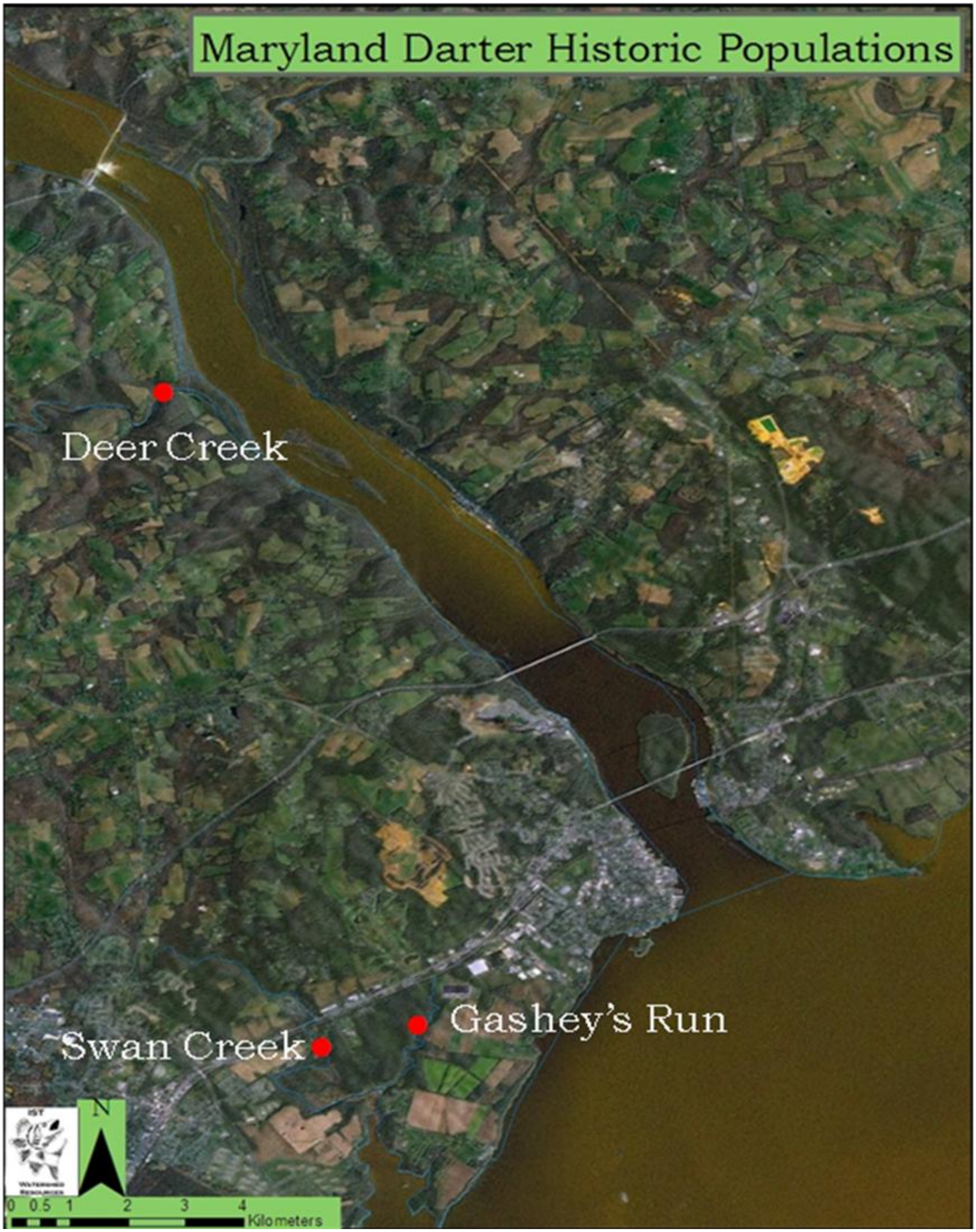


Figure 2: Historic locations of Maryland darter collections.

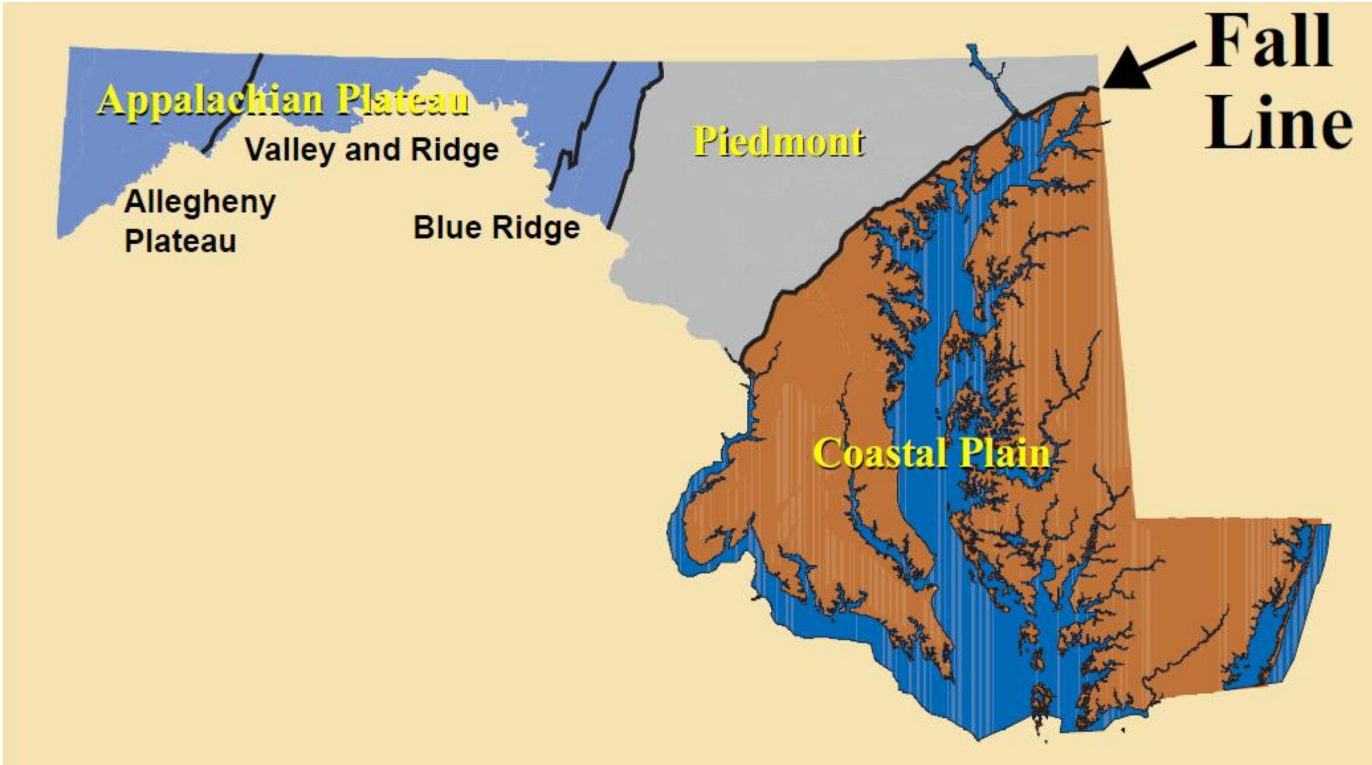


Figure 3: The Fall Line. Map modified from Boward et al. 1999.

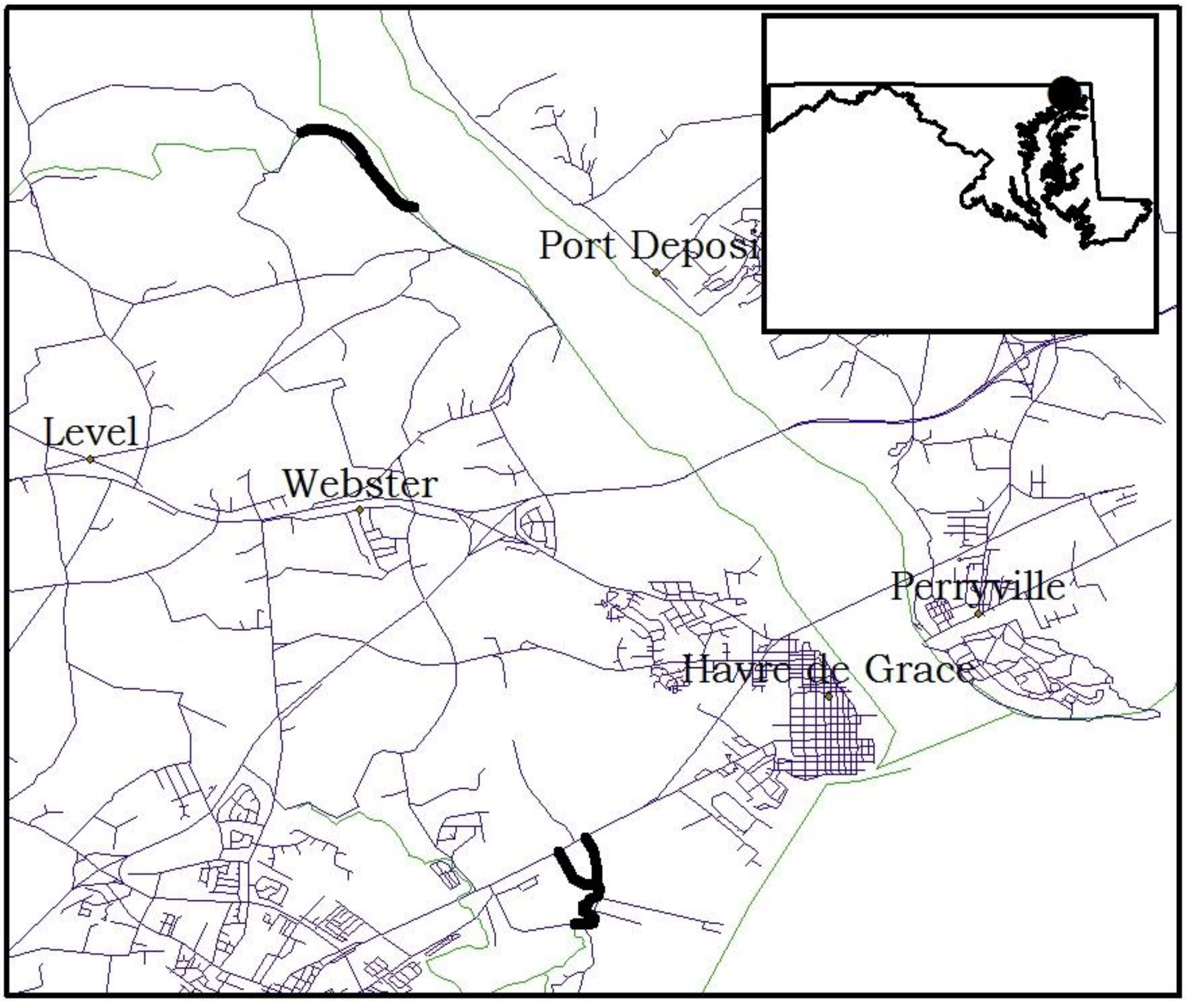


Figure 4: Established Critical Habitat for *Etheostoma sellare* map. (a recreation of map within U.S. Fish and Wildlife Service 1984)

Figure 5: Susquehanna mainstem below the Conowingo dam. (Note: large areas of exposed riverbed)

Figure 6: Deer Creek electro-fishing sample reach.

Figure 7: Swan Creek electro-fishing sample reach.

Figure 8: Gashey’s Run electro-fishing sample reach.

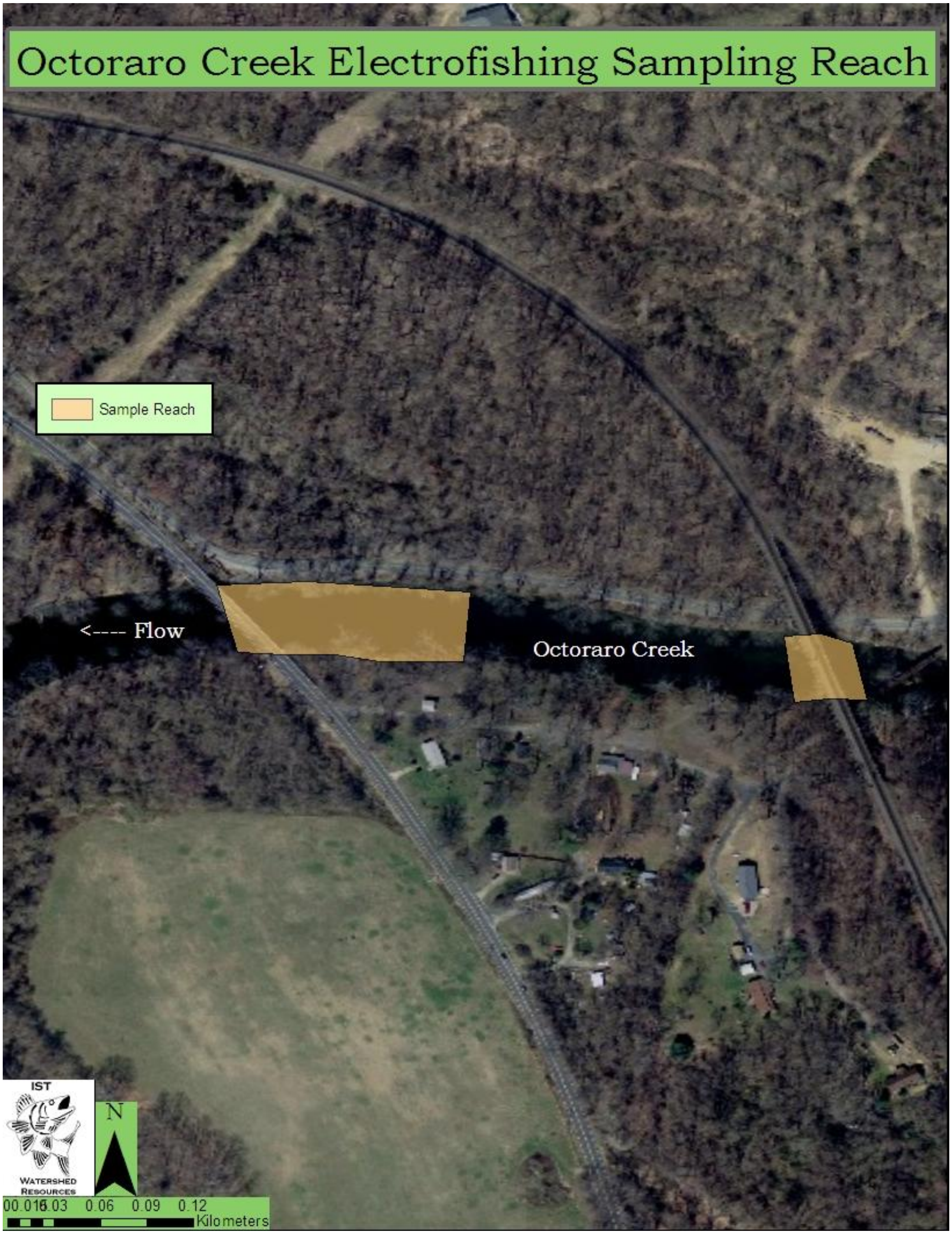


Figure 9: Octoraro Creek electro-fishing sample reach.

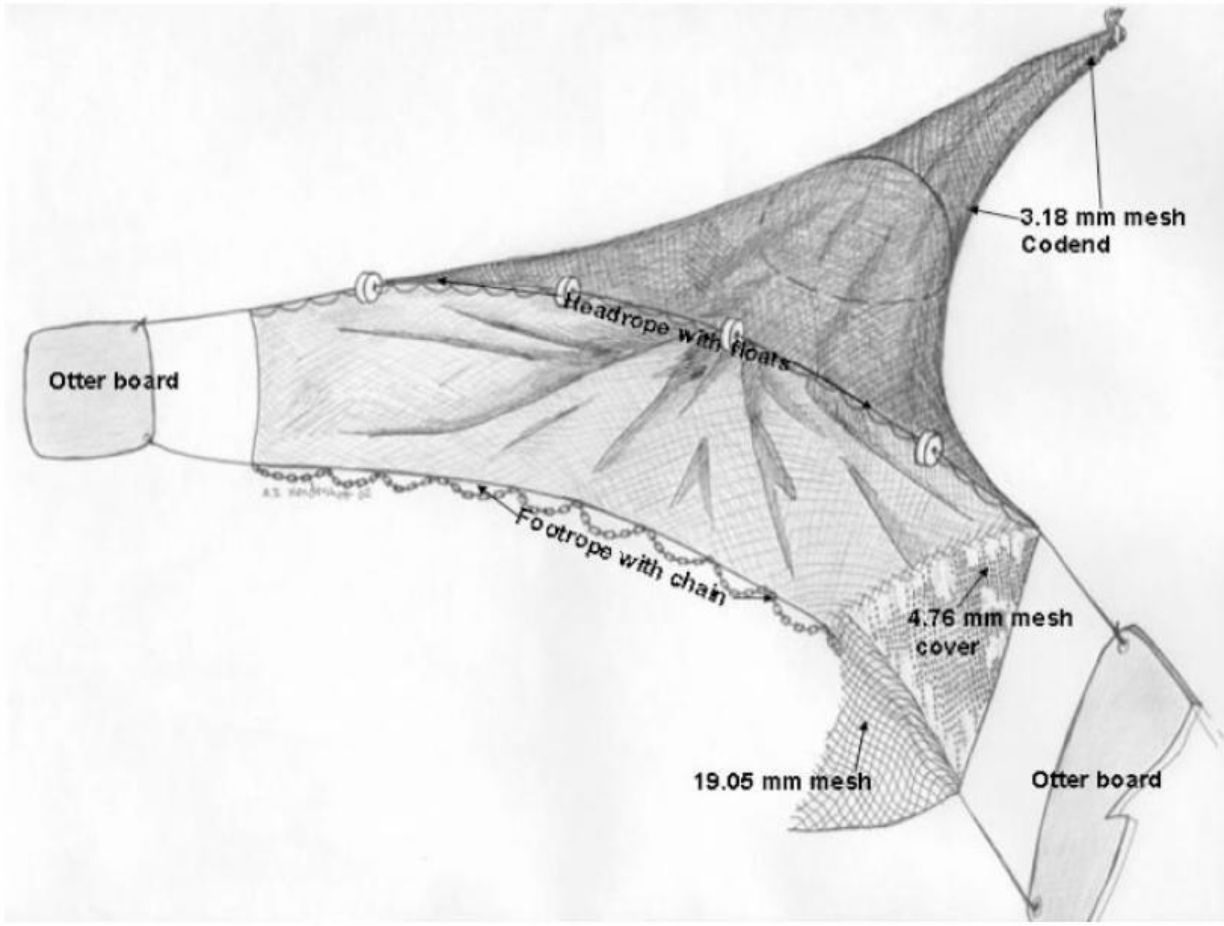


Figure 10: Illustration Modified Missouri trawl.

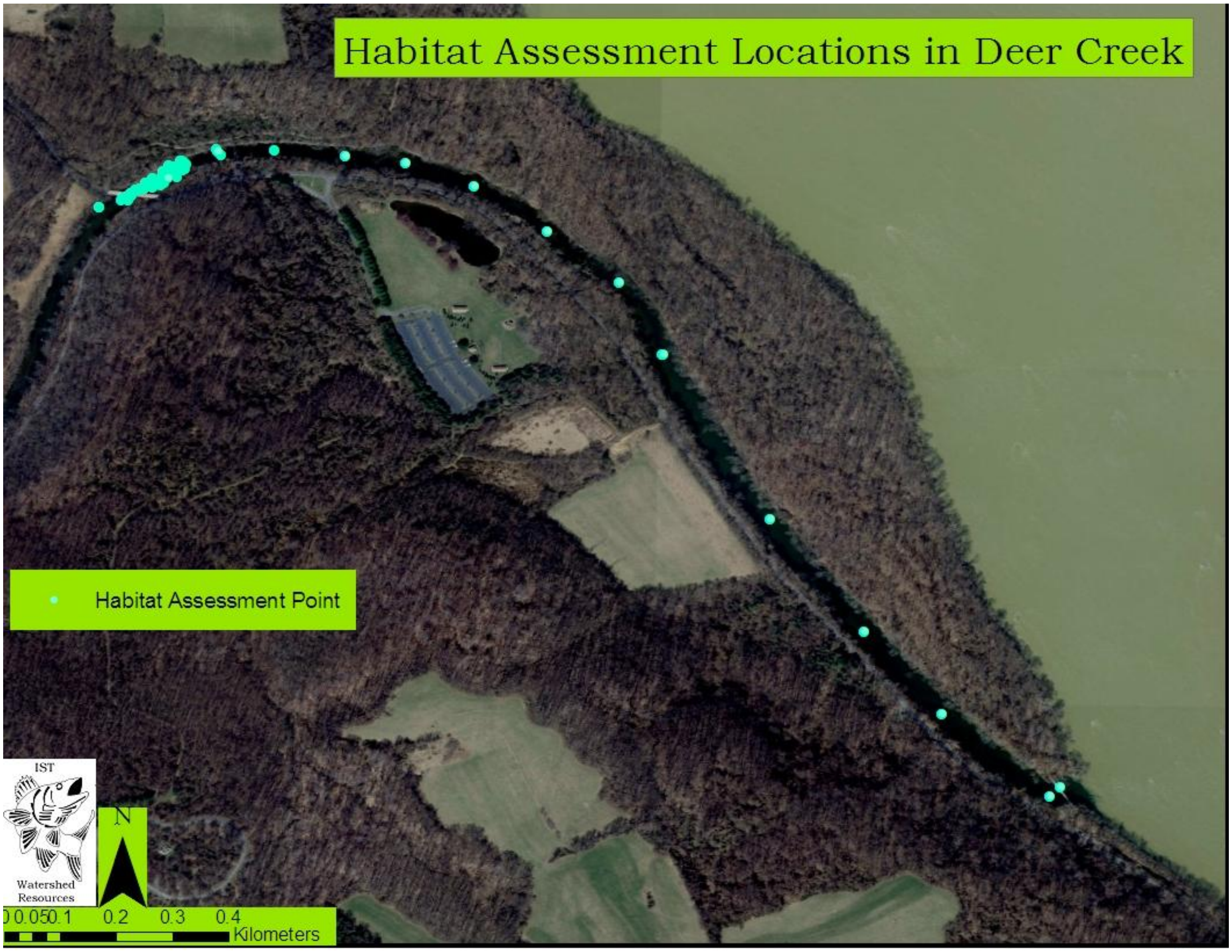


Figure 11: Habitat assessment points in Deer Creek.

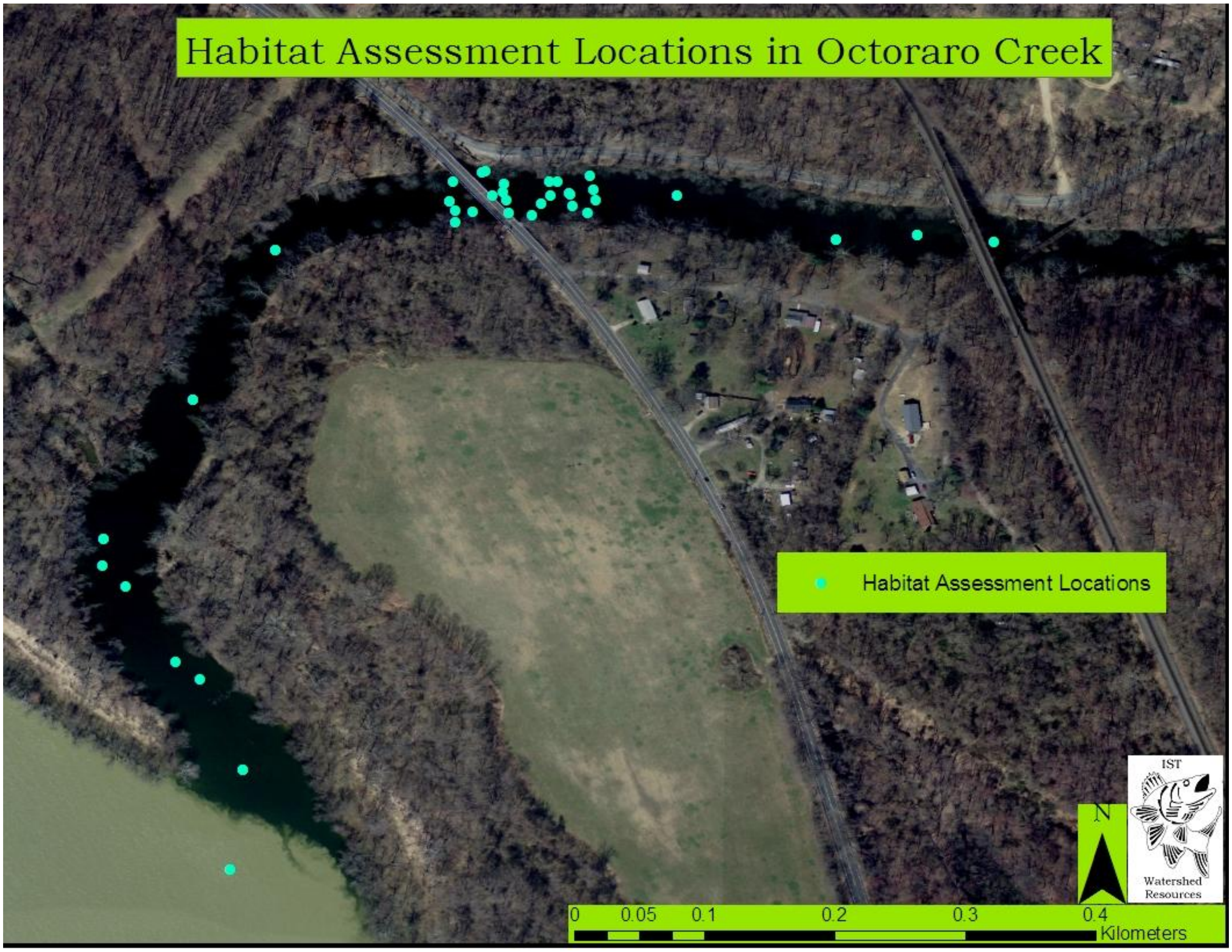


Figure 12: Habitat assessment points in Octoraro Creek

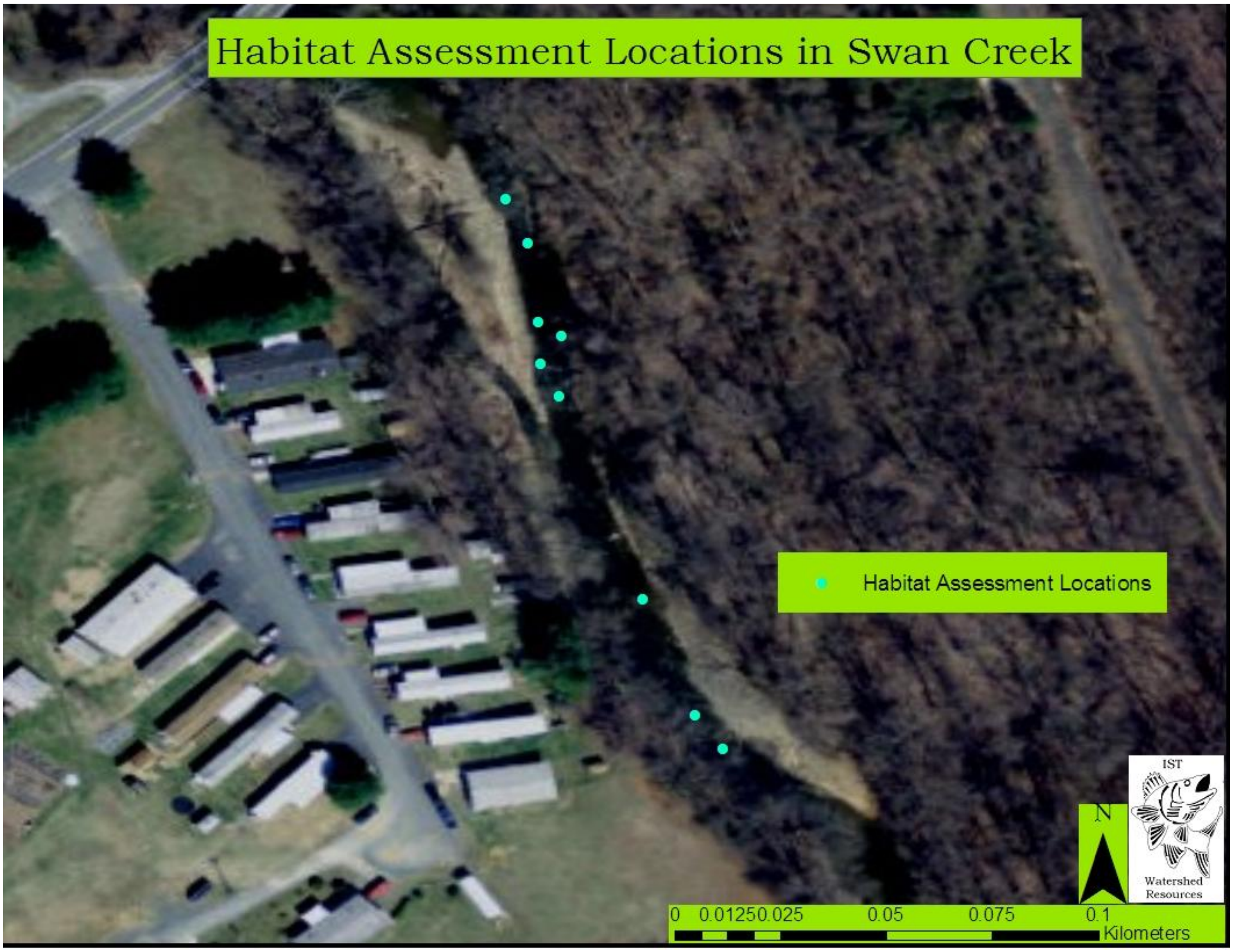


Figure 13: Habitat assessment points in Swan Creek

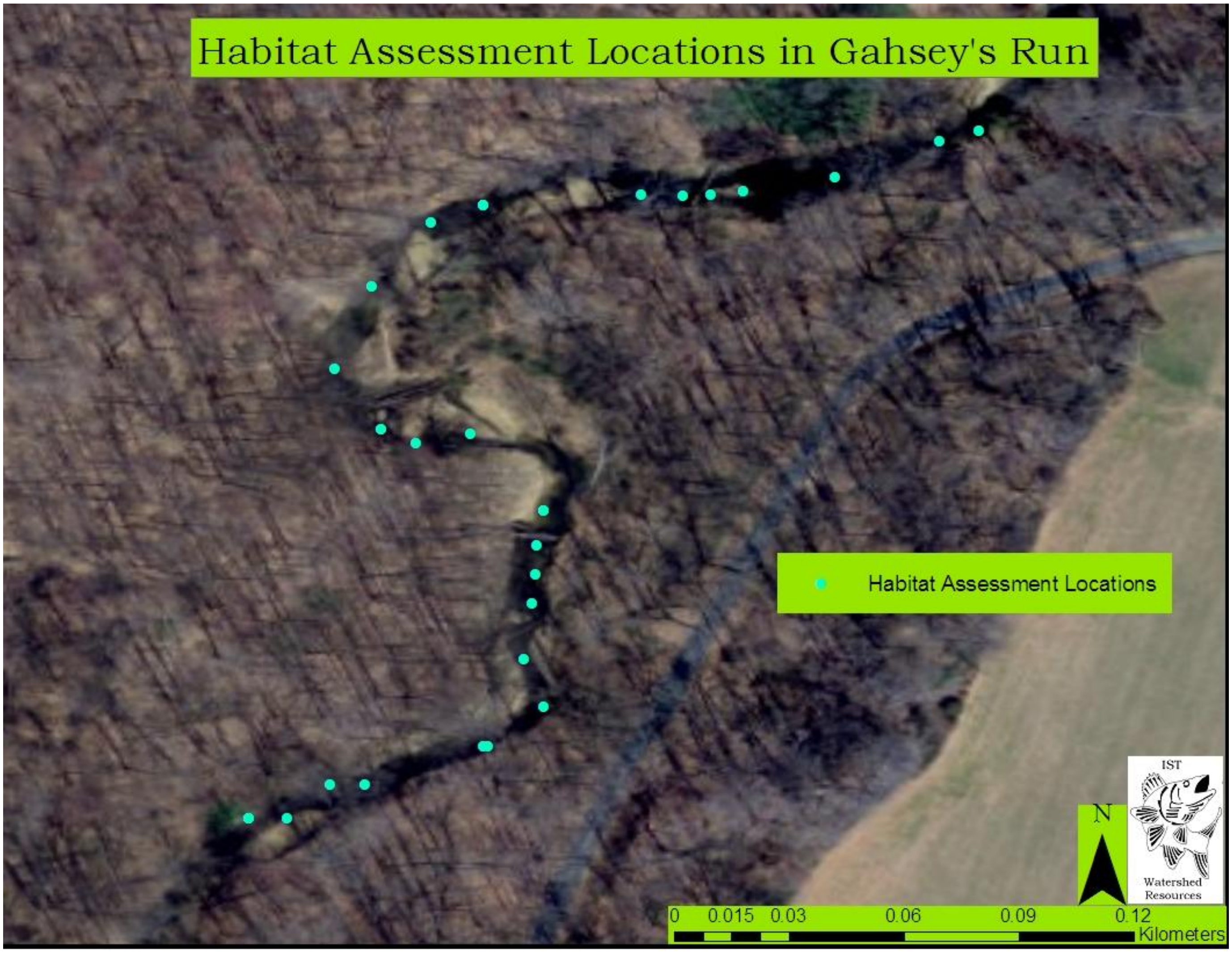


Figure 14: Habitat assessment points in Gashey's Run

Visual Survey Scales

Relative Velocity
1-low
2-moderate
3-high

Dominant Substrate
1-fines
2-sand
3-gravel
4-cobble
5-boulder
6-bedrock

Algae Coverage
0-none
1-patchy
2-full coverage
3-elevated growth

Fish Abundance
0-none
1-one to four
2-five to ten
3-ten to twenty
4->20

Snail Abundance
0-none
1-scattered individuals
2-abundant
3-complete coverage

Mussel Abundance
0- No live No Dead
1- Dead shells only or 1-5 live
2- 6-10 live mussels
3- 10-20 live mussels
4- >20 live mussels

Figure 15: Visual Survey Scales.

Figure 16: Area covered by visual surveys at Deer Creek Stafford Bridge crossing location.

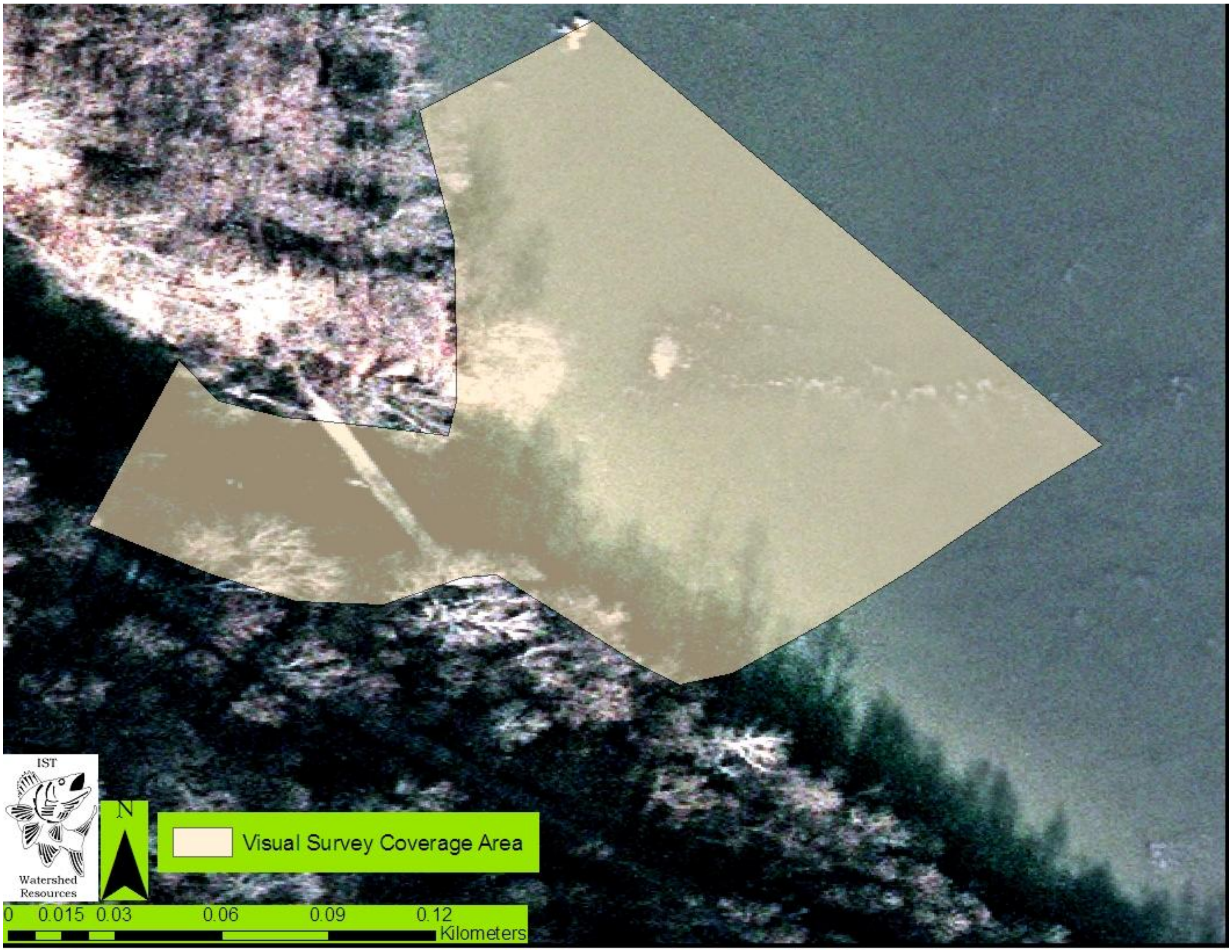
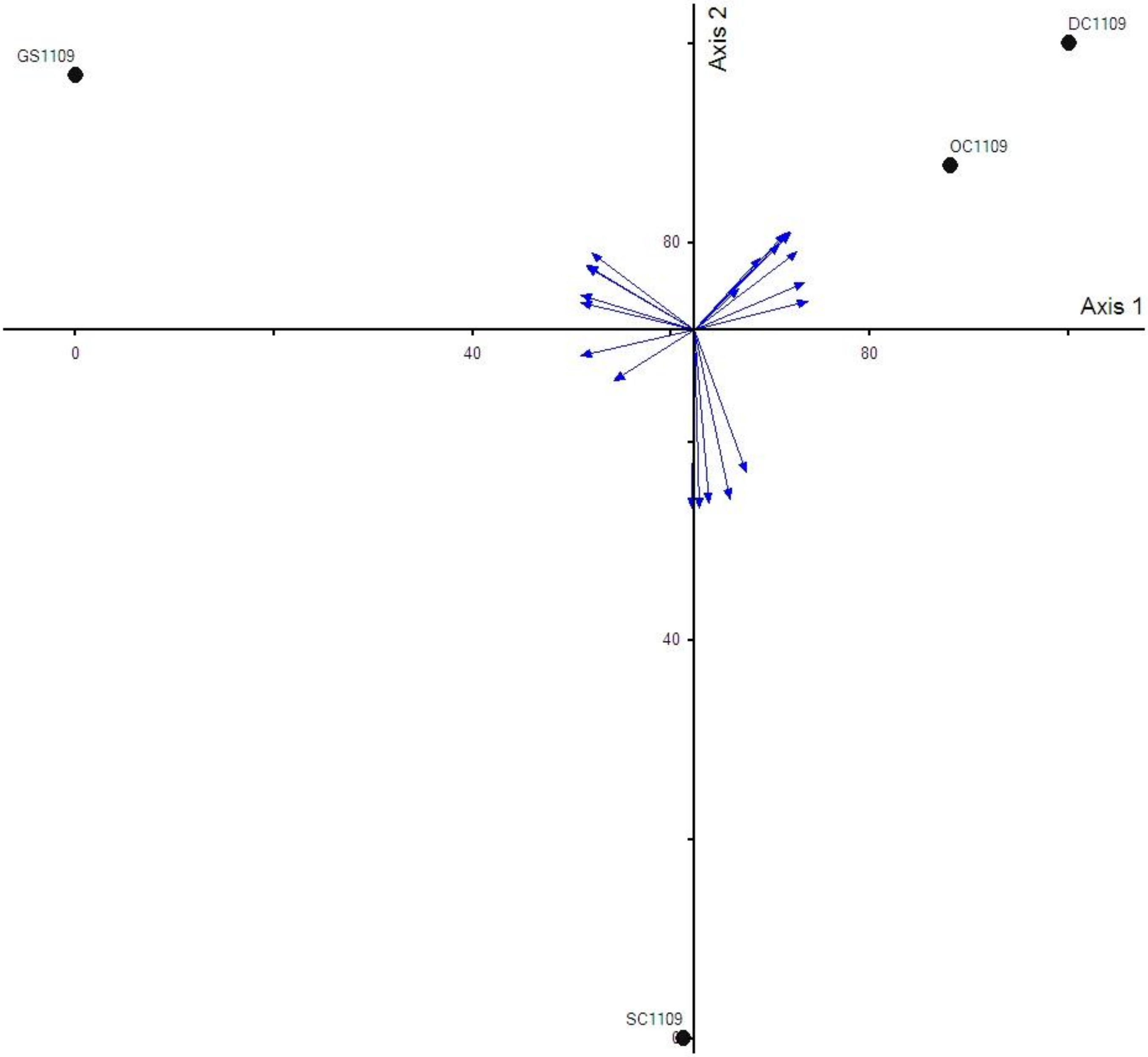


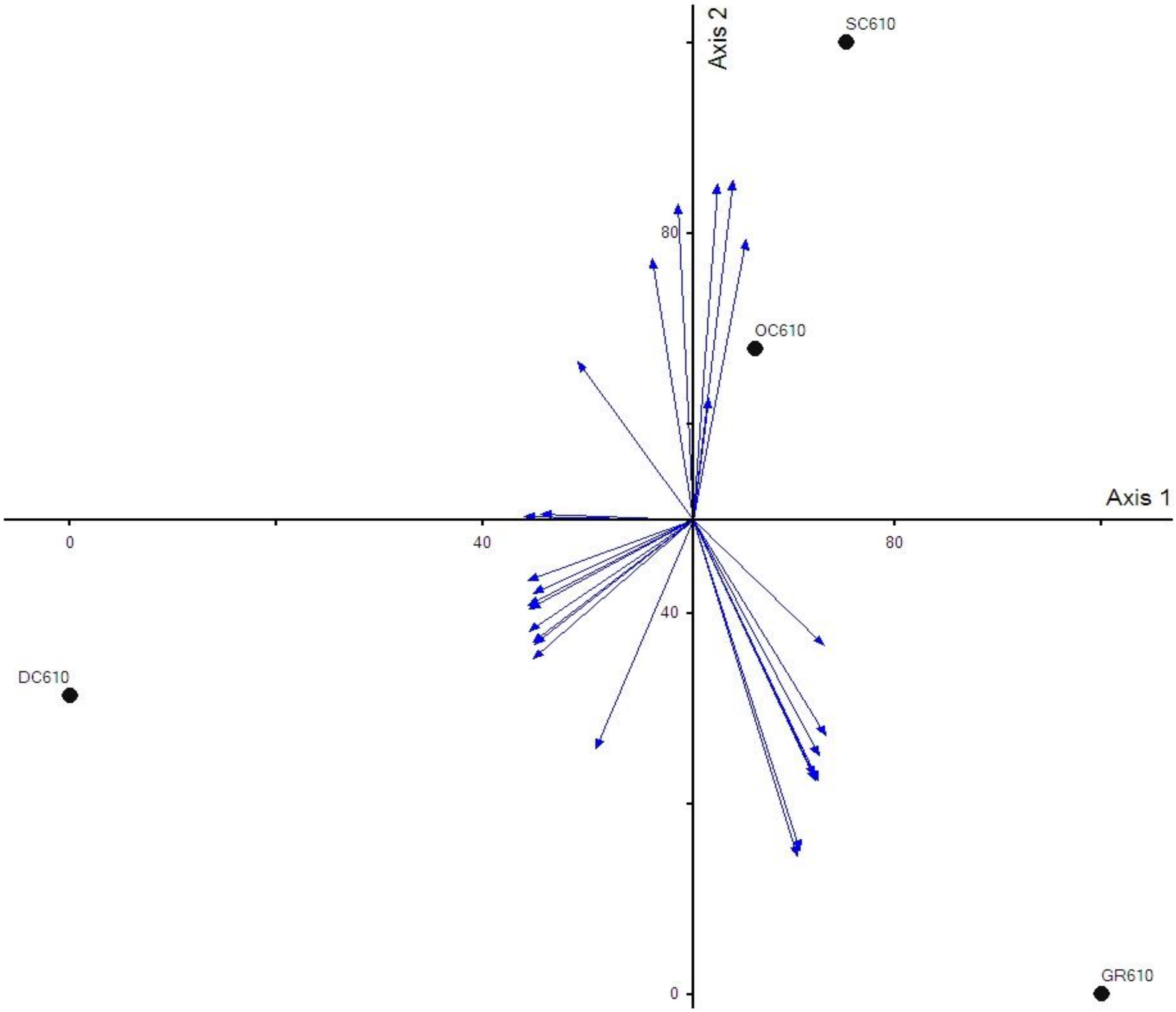
Figure 17: Area covered by visual surveys at the mouth of Deer Creek.

Figure 18: Area covered by visual surveys at Octoraro Creek.



AXIS	Eigenvalue	% of Variance	Cum.% of Var.	Broken-Stick Eigenvalue
1	14.440	53.481	53.481	3.891
2	10.044	37.200	90.681	2.891
3	2.516	9.319	100.00	2.391
4	0.000	0.000	100.00	1.808
5	0.000	0.000	100.00	1.608

Figure 19: PCA of the 2009 electro-fishing data grouped by site.



AXIS	Eigenvalue	% of Variance	Cum.% of Var.	Broken-Stick Eigenvalue
1	20.058	60.781	60.781	4.089
2	7.237	21.930	82.711	3.089
3	5.705	17.289	100.00	2.589
4	0.000	0.000	100.00	2.255
5	0.000	0.000	100.00	2.005

Figure 20: PCA of June 2010 electro-fishing data grouped by site.

Figure 21: Map of mainstem visual survey sub-reach locations.

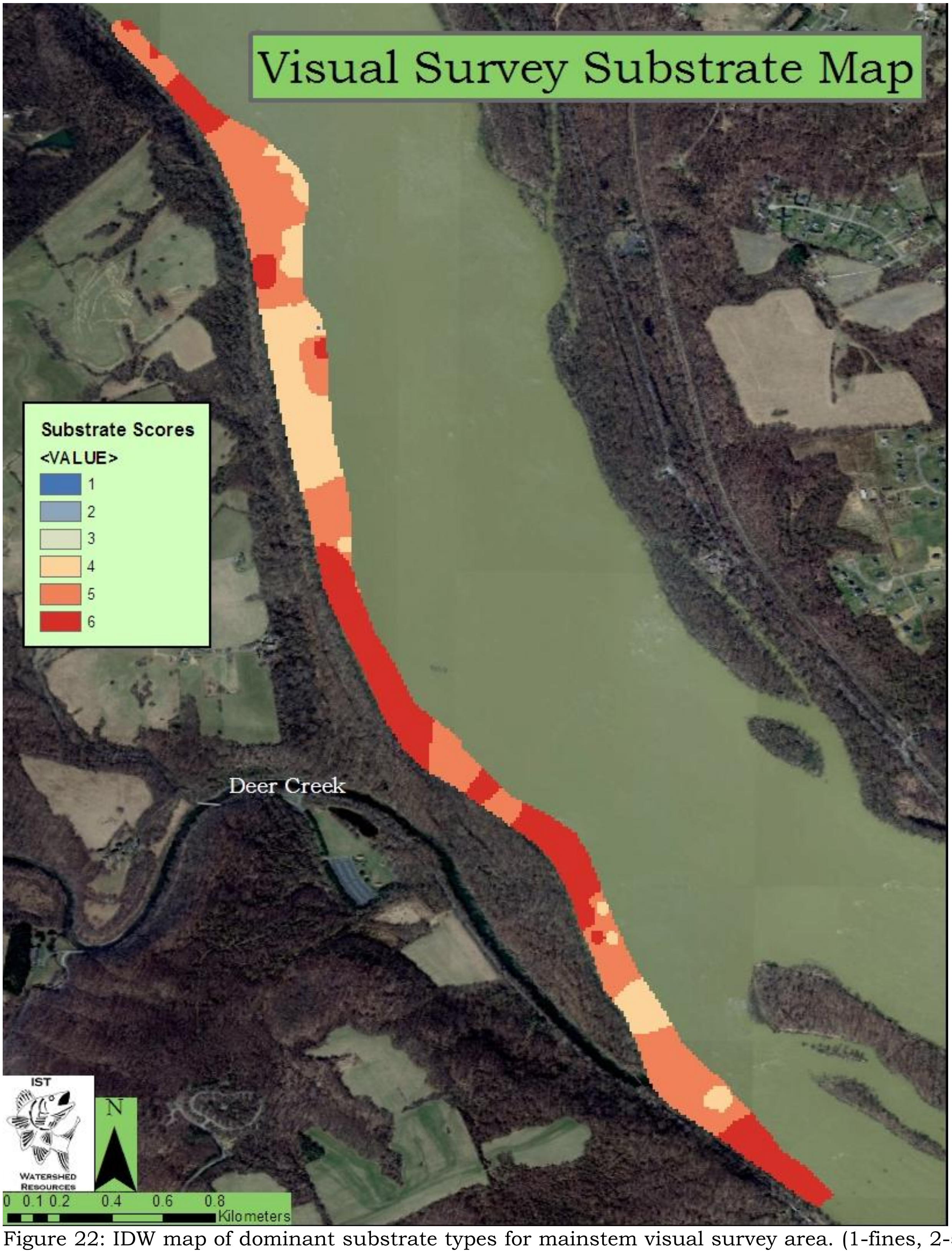


Figure 22: IDW map of dominant substrate types for mainstem visual survey area. (1-fines, 2-sand, 3-gravel, 4-cobbel, 5-boulder, and 6-bedrock)

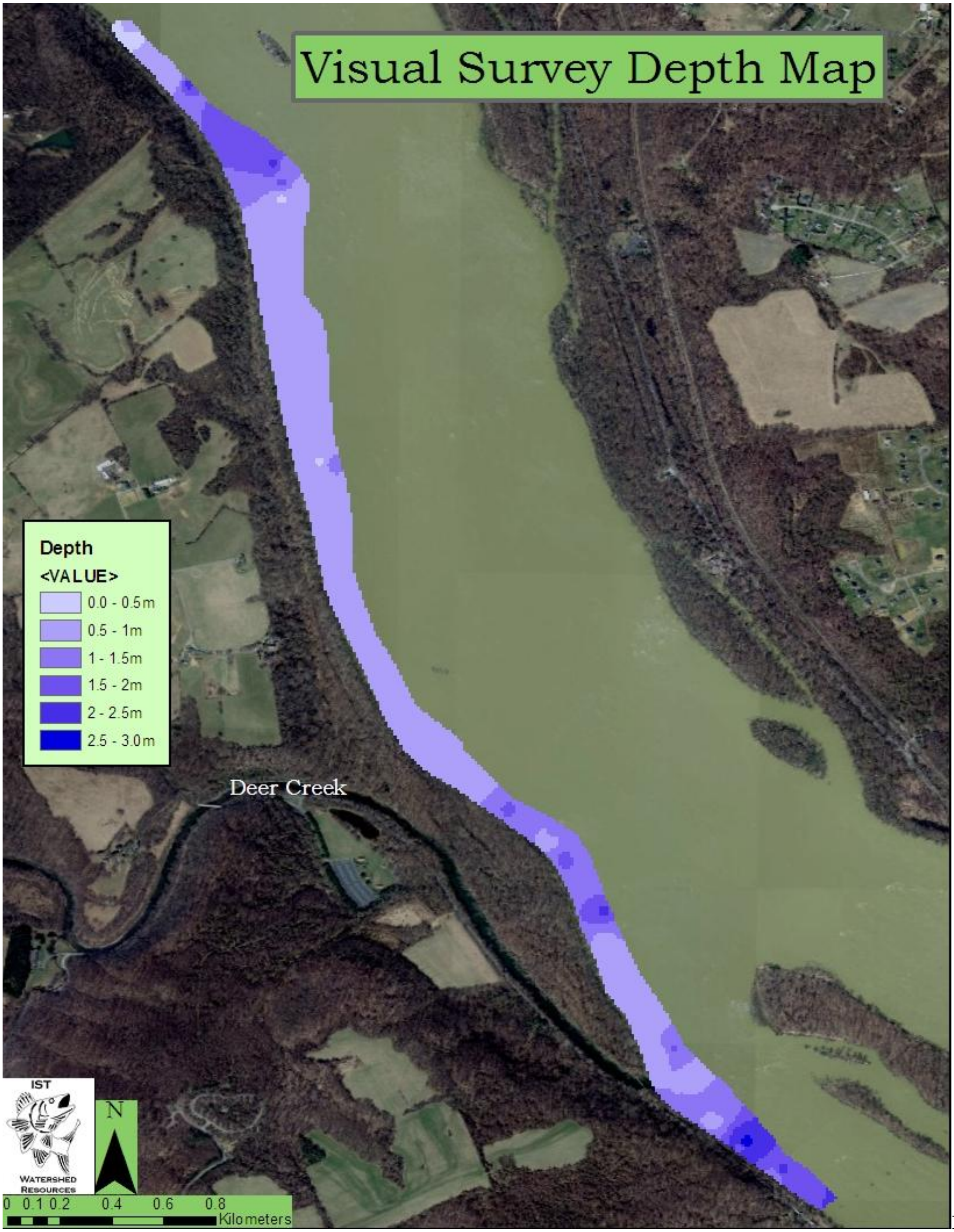


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Figure 23: IDW map of depth for the mainstem visual survey area.

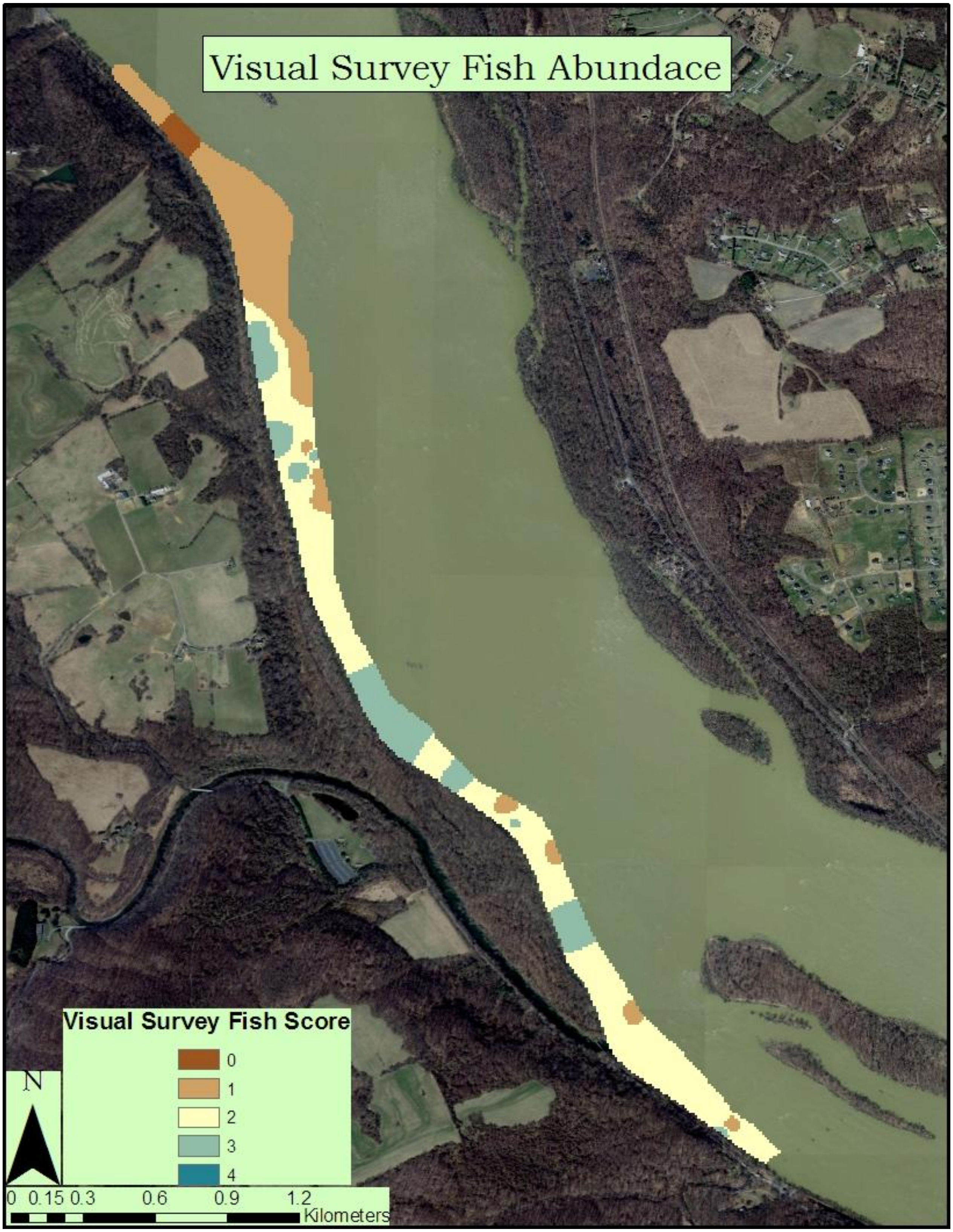

Figure 24: IDW map of fish scores for the mainstem visual survey area. (0=no fish, 1=1-4 individuals , 2=5-10 individuals , 3=10-20 individuals, and 4=>20 individuals)

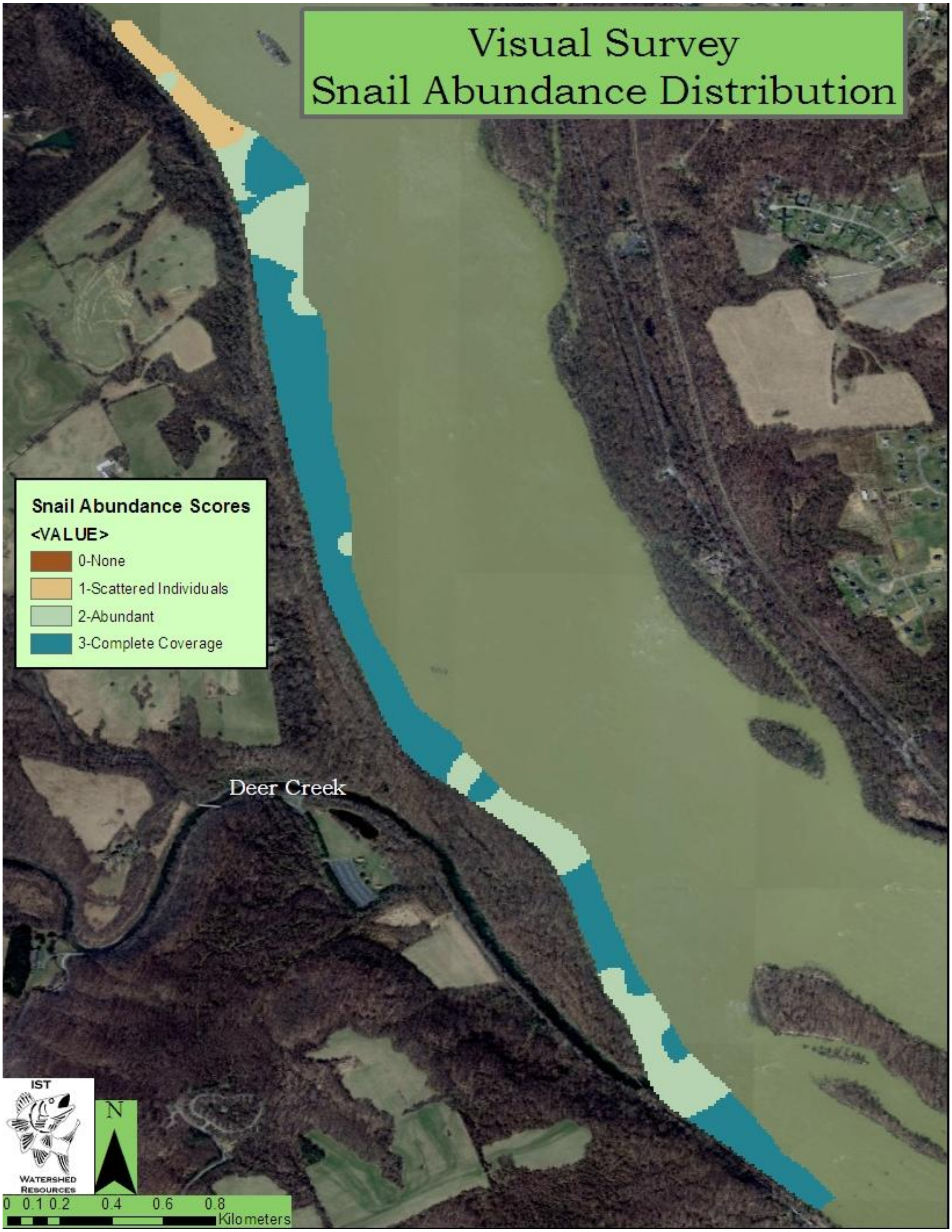

Figure 25: IDW map of snail density for the mainstem visual survey area.

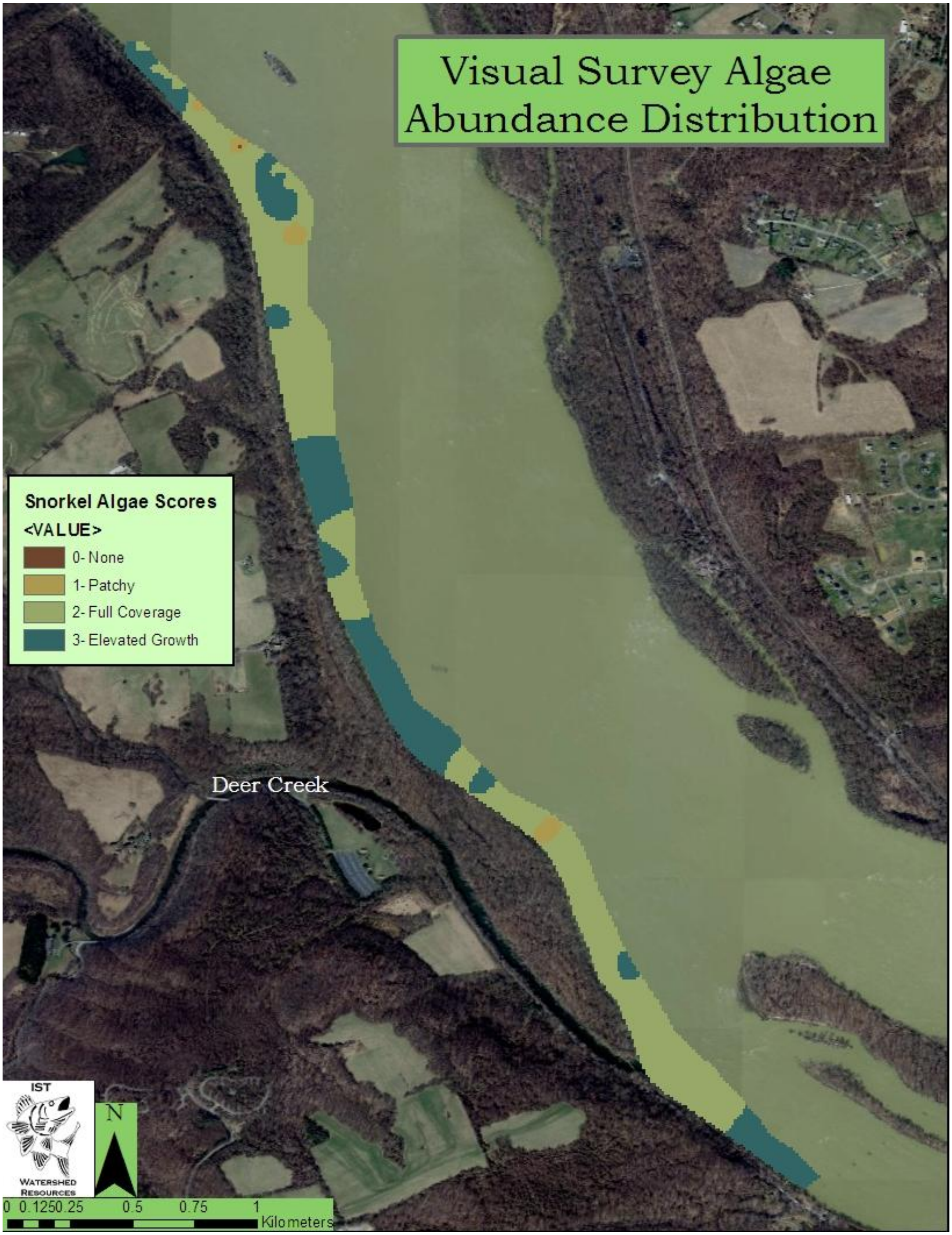

Figure 26: IDW map of algae coverage for the mainstem visual survey area.

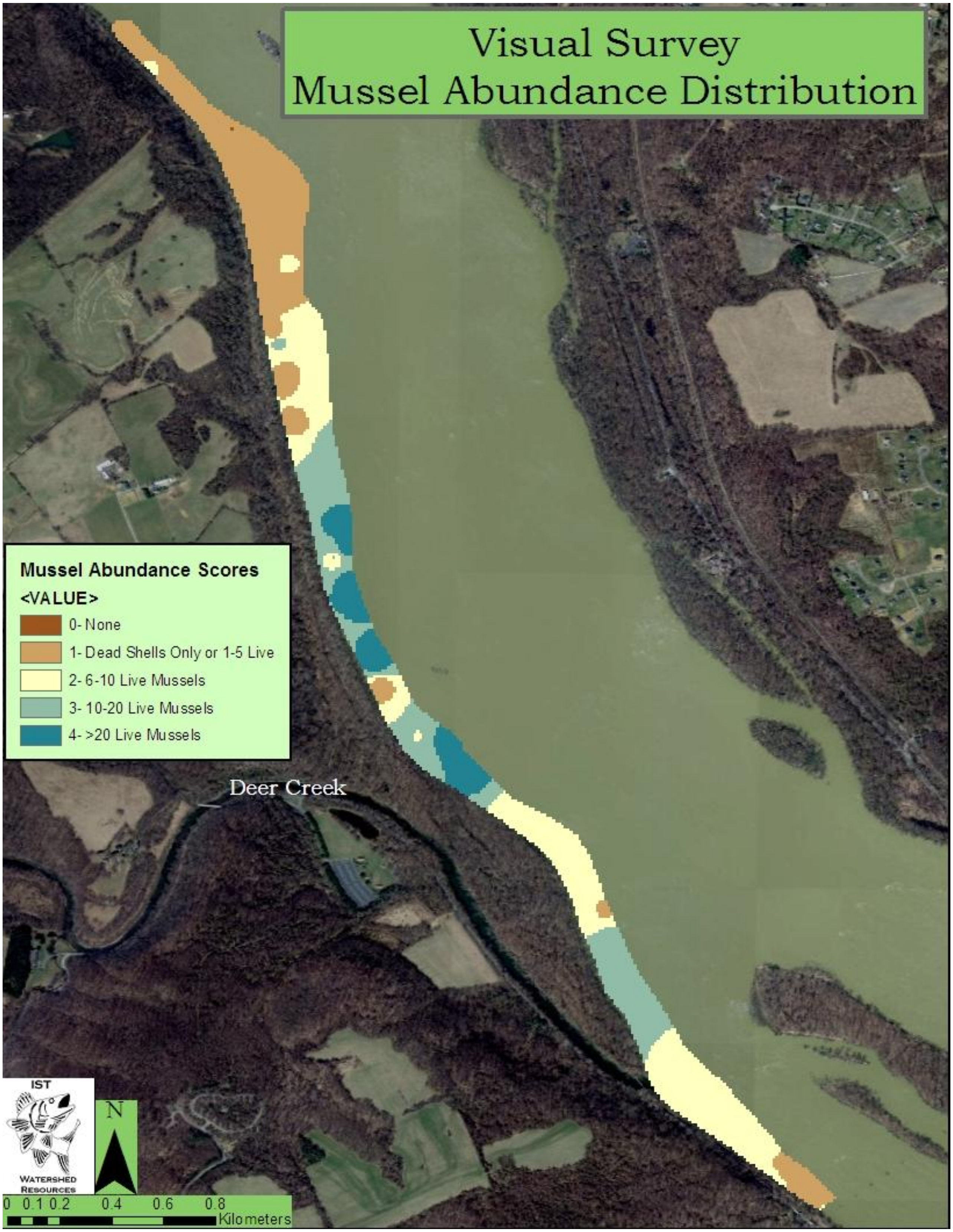

Figure 27: IDW map of mussel density for the mainstem visual survey area.

Figure 28: Map showing observed areas of silt/sand deposits at the mouth of Swan Creek.

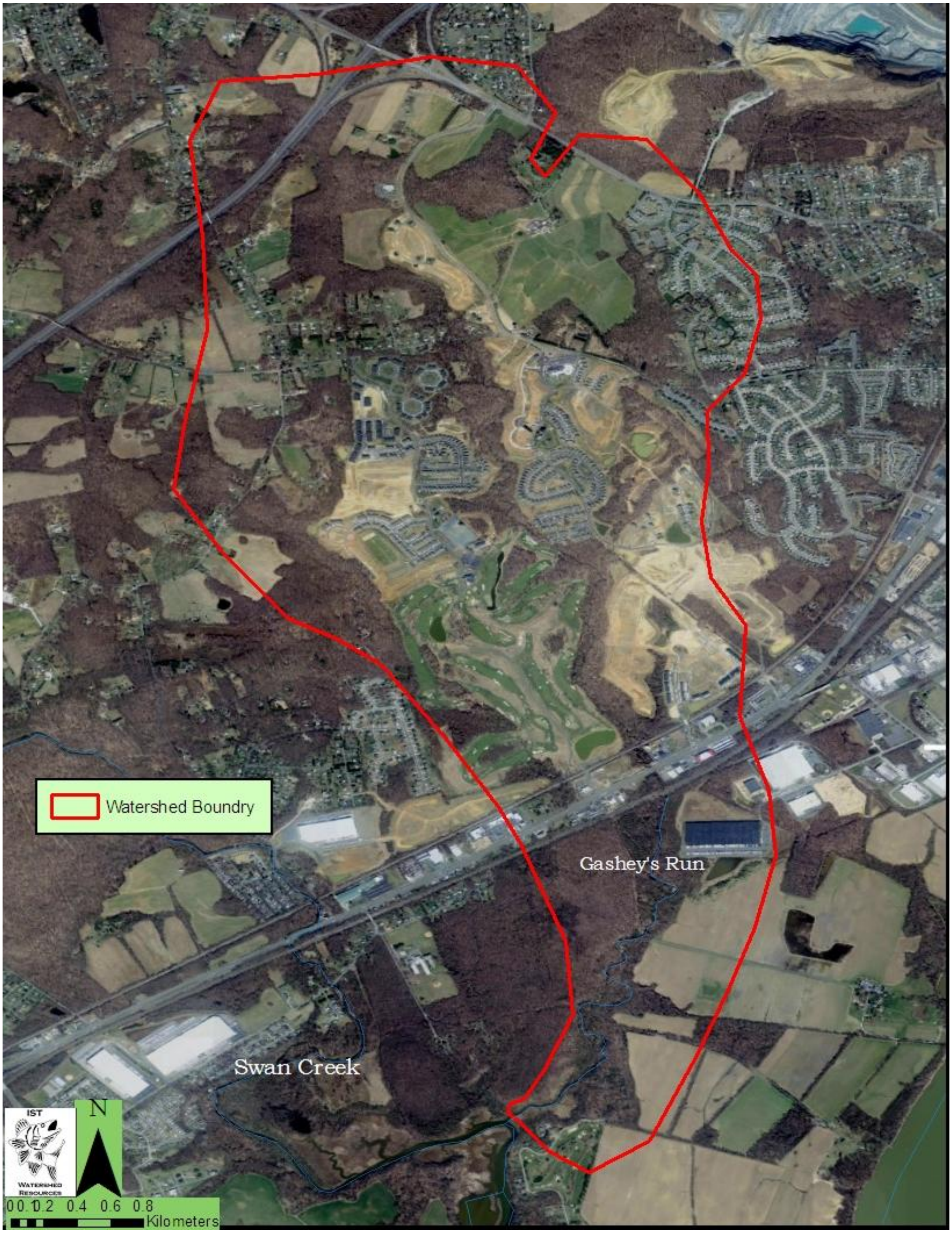

Figure 29: Map showing large areas of urbanization occurring in the Gashey's Run watershed.

Figure 30: Picture showing examples of erosion at the Gashey's Run Site. Photo by Tyler Hern

Figure 31: Picture showing examples of erosion at the Gashey's Run Site. Photo by Nathan Hoxie

Figure 32: Picture showing Gashey's Run during a low flow event in July. Photo by Nathan Hoxie

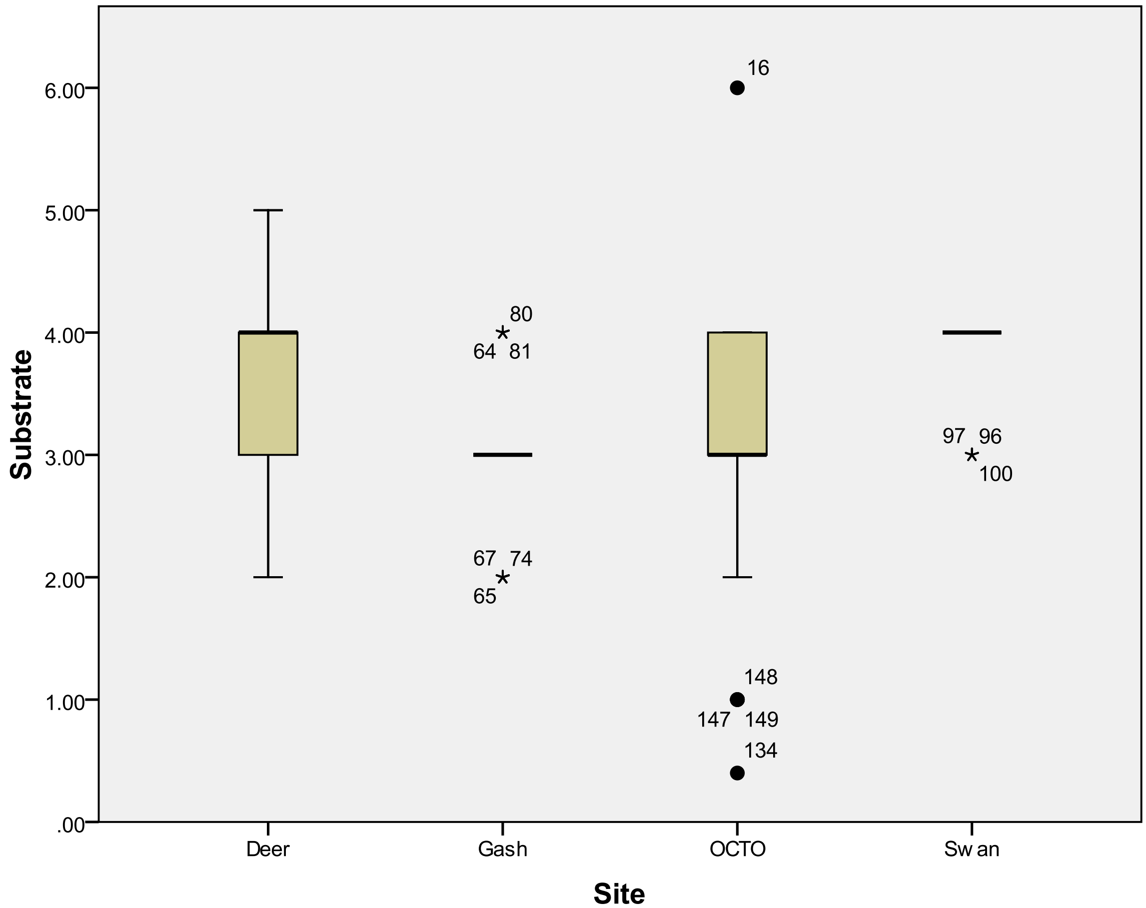

Figure 33: Box and Whisker plot of substrate data taken from stream sites.

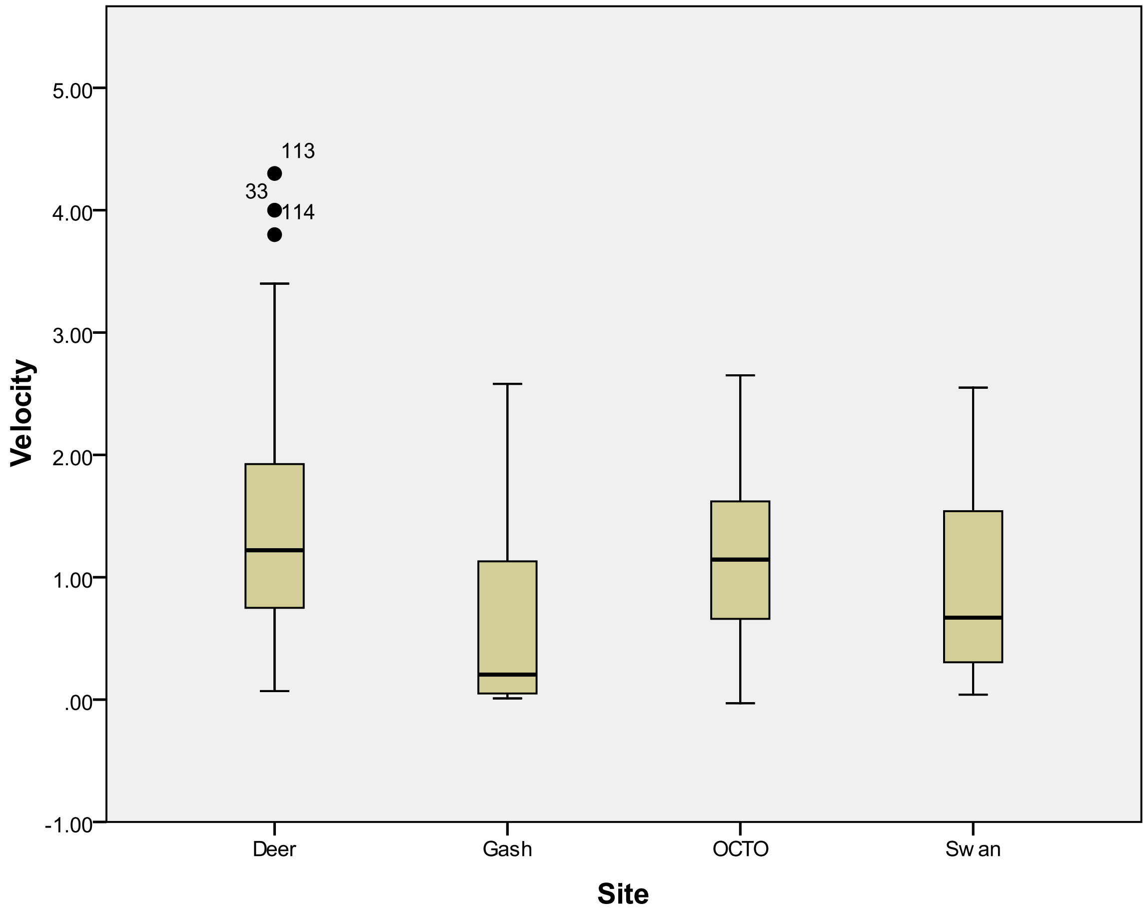

Figure 34: Box and Whisker plot of velocity data taken from stream sites.

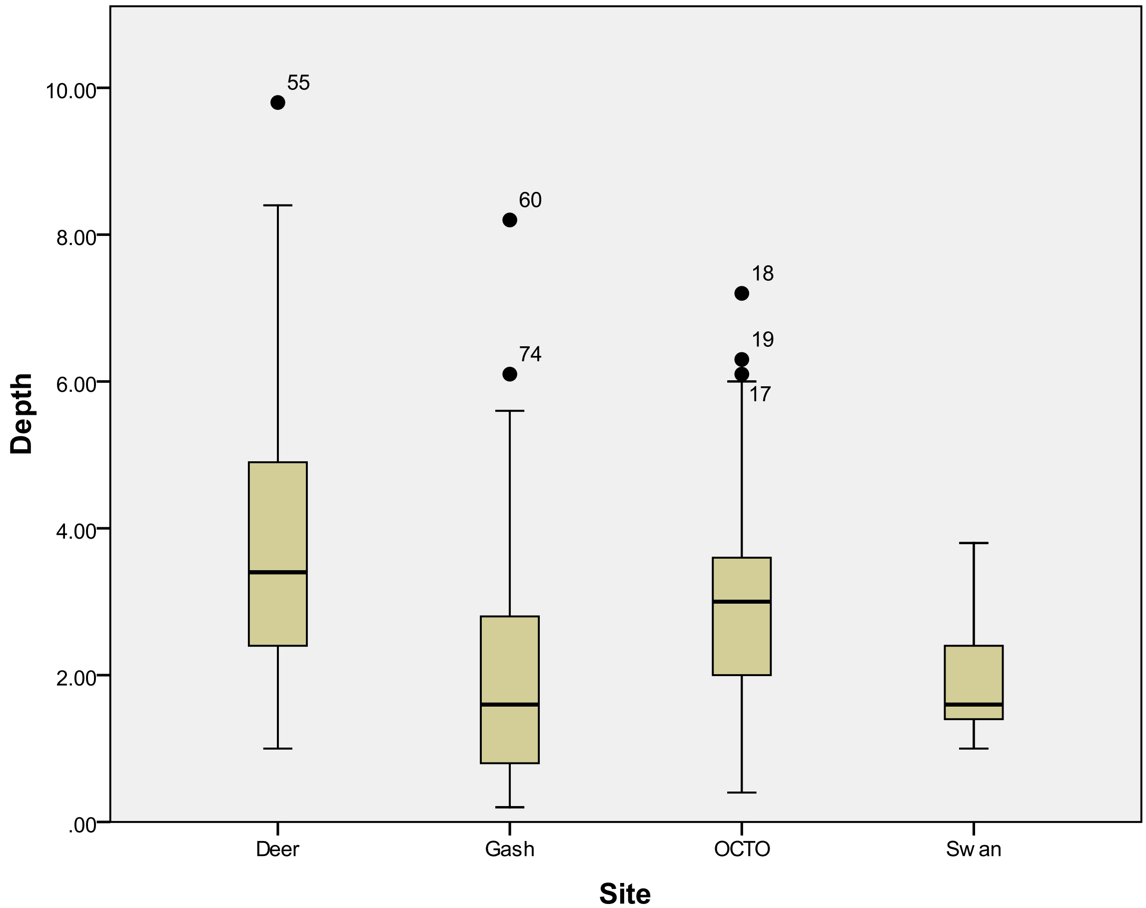

Figure 35: Box and Whisker plot of depth data taken from stream sites.

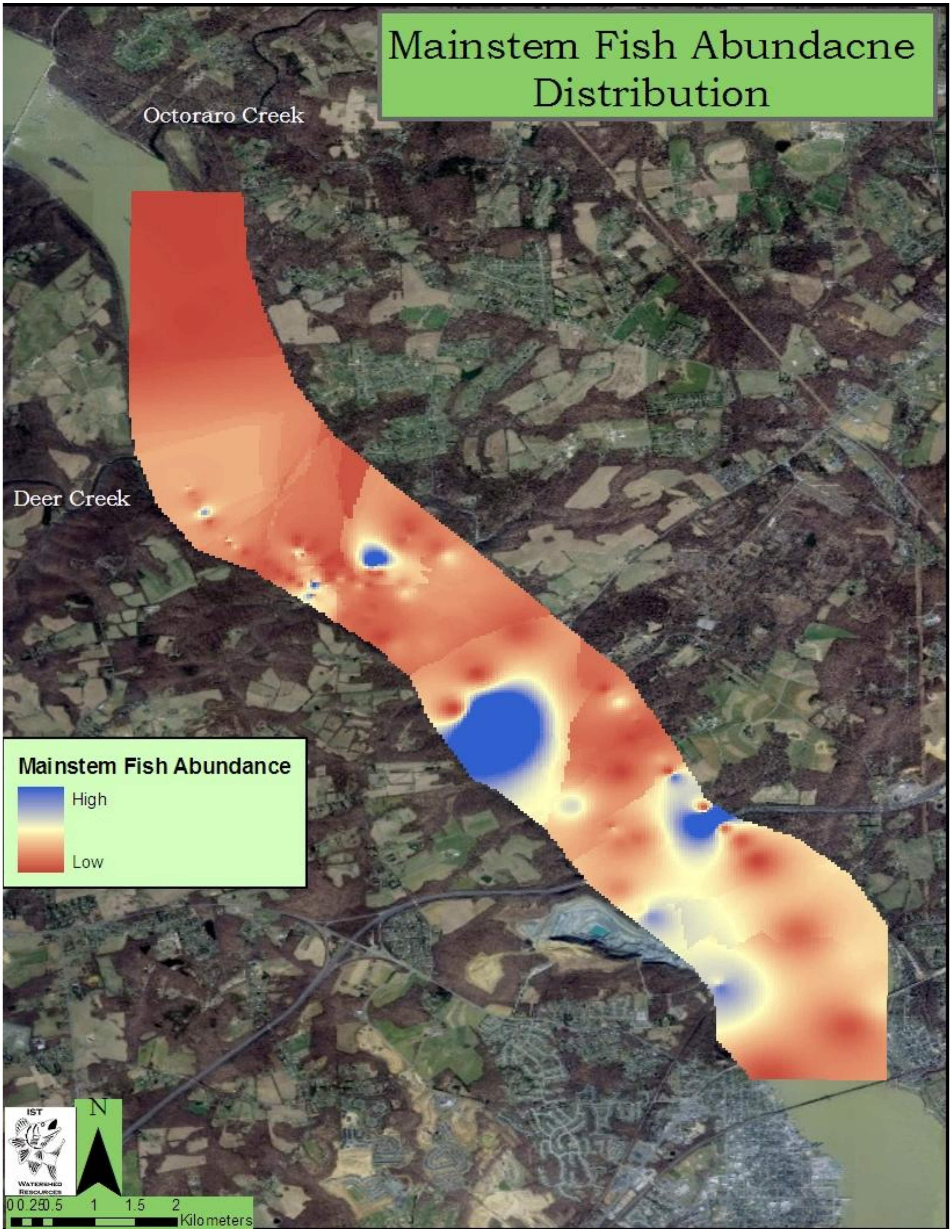

Figure 36: IDW map showing fish abundance taken from trawling data 2008 and 2009.

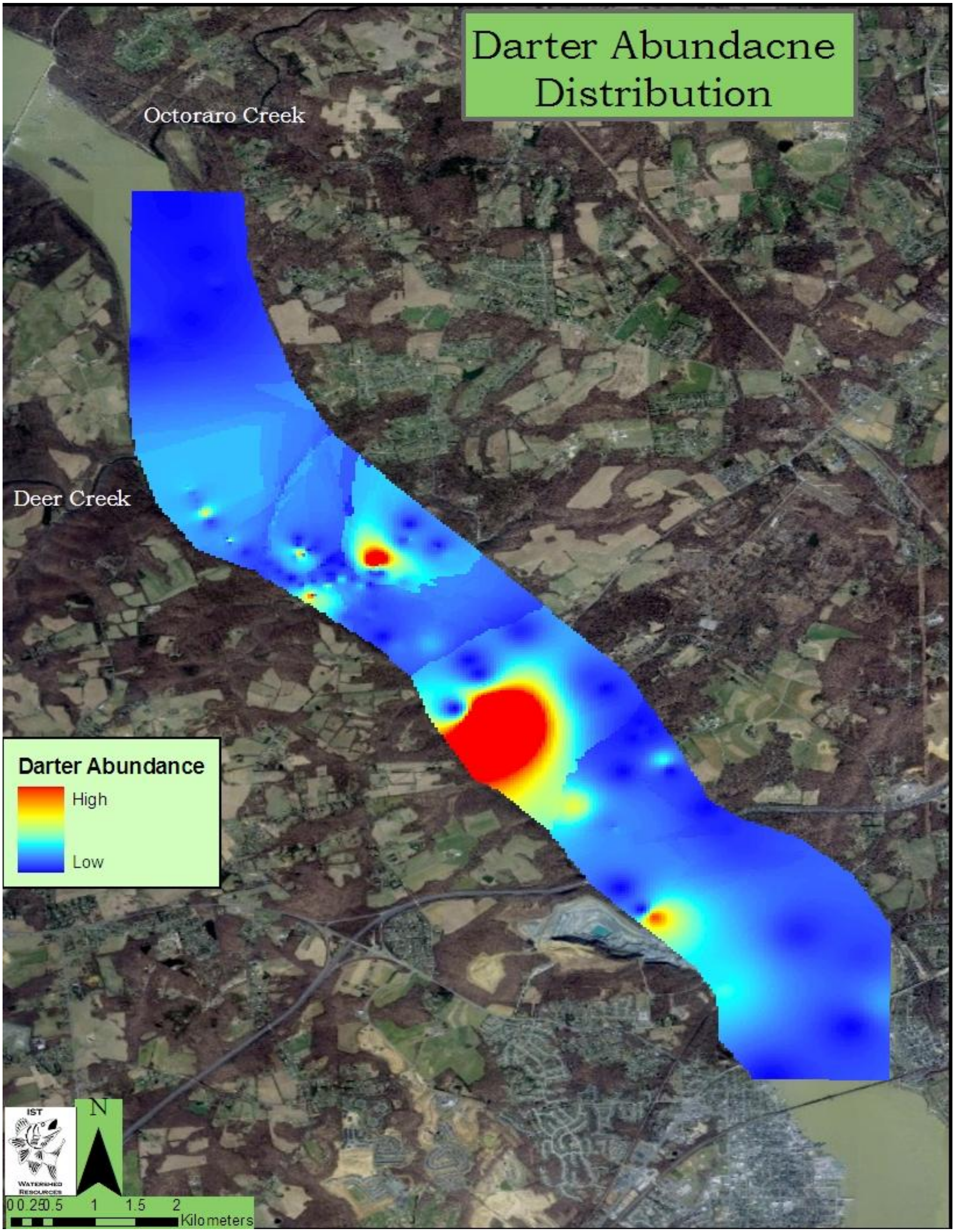

Figure 37: IDW map showing darter abundance taken from trawling data 2008 and 2009.

Figure 38: Map showing area of mainstem with similar habitat attributes to the Deer Creek site.

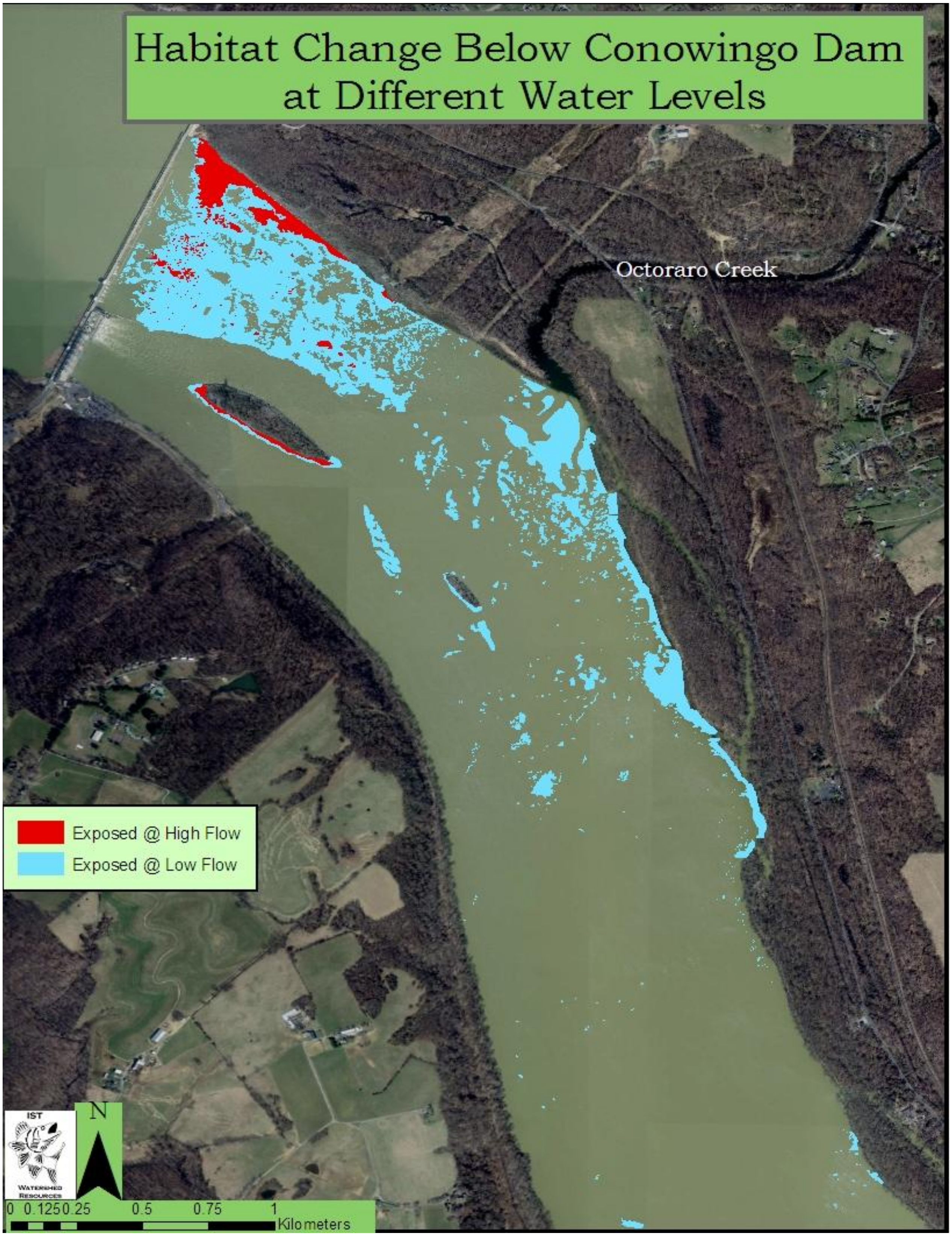

Figure 39: Map showing change in exposed river bed during high and low flow events below the Conowingo Dam.

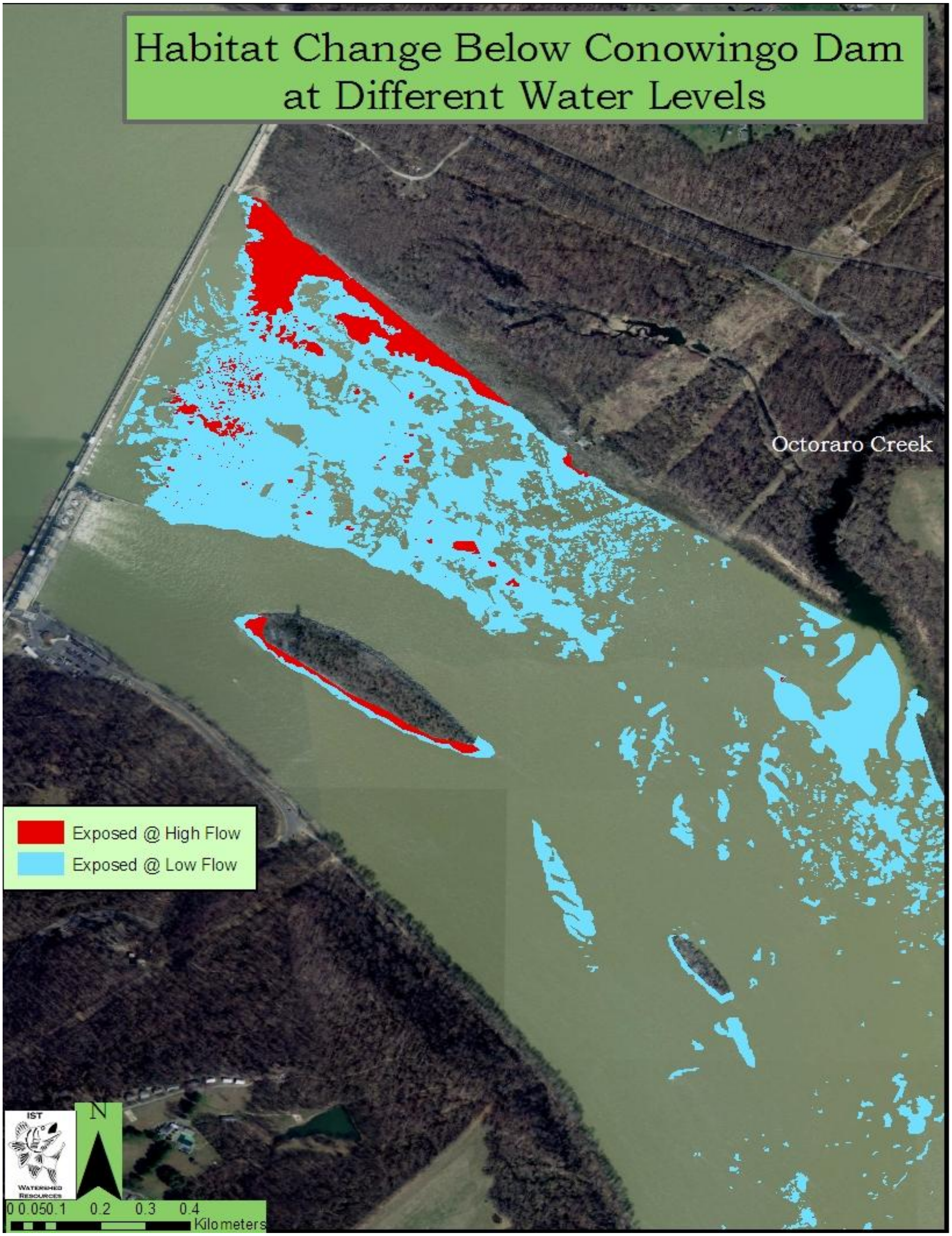

Figure 40: Map showing change in exposed river bed at the mouth of Octoraro Creek during high and low flow events below the Conowingo Dam.

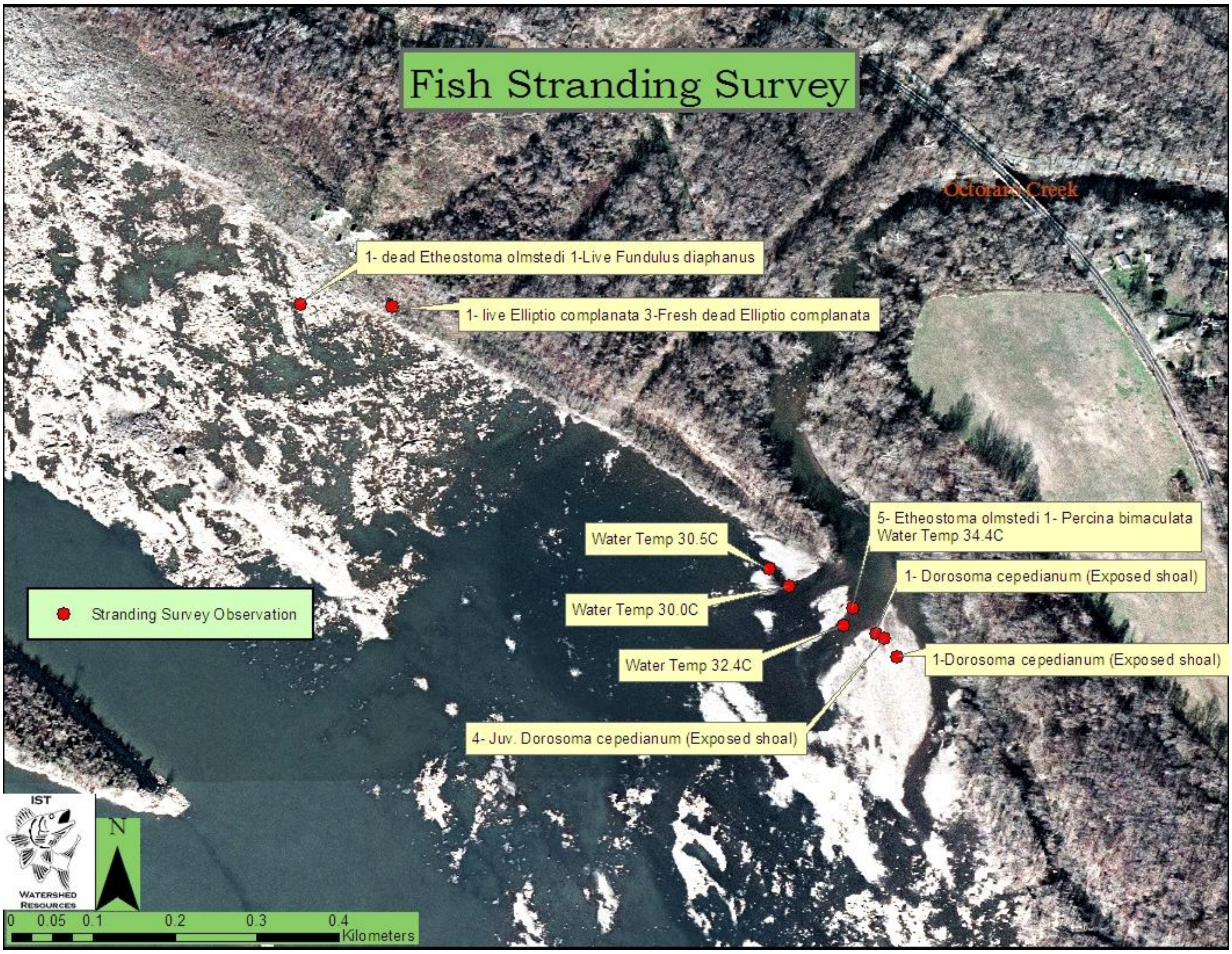

Figure 41: Map showing the locations of observed stranded fish and temperatures of side channels at a low flow event July, 8 2010.

Figure 42: Map showing the locations of snail collections in Deer Creek.

Figure 43: Map showing the locations of snail collections in Octoraro Creek.

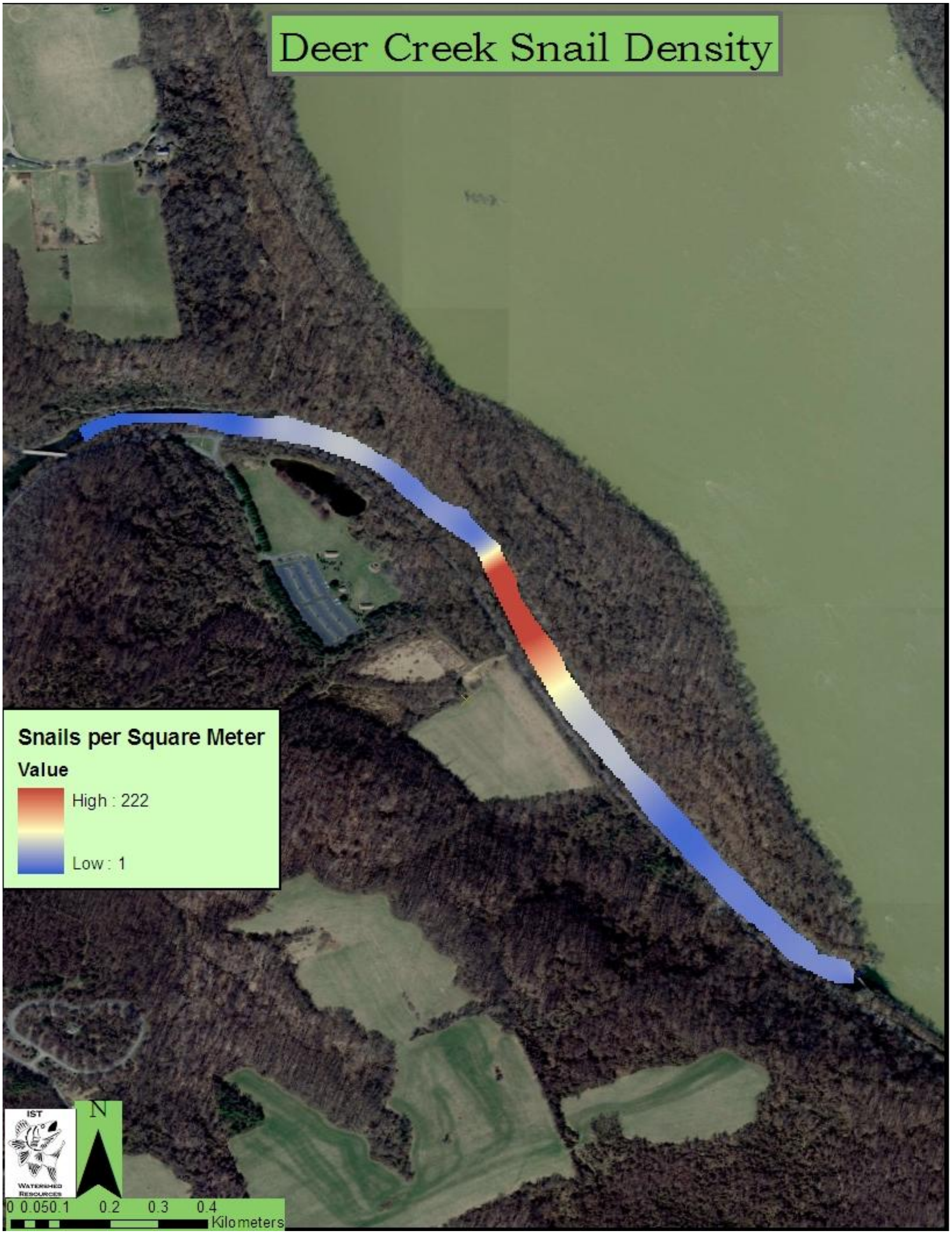

Figure 44: Snail density distribution in Deer Creek.

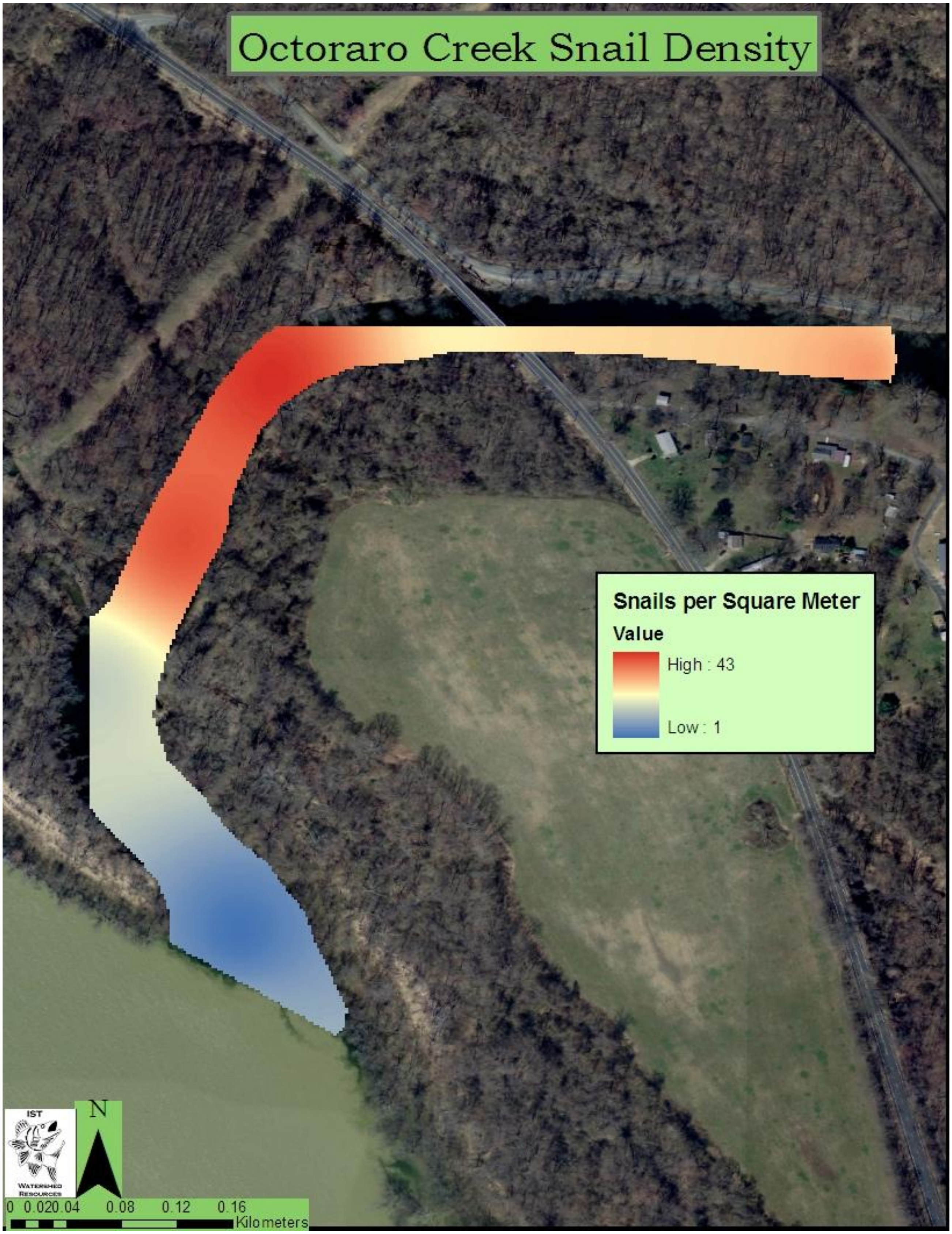

Figure 45: Snail density distribution in Octoraro Creek.

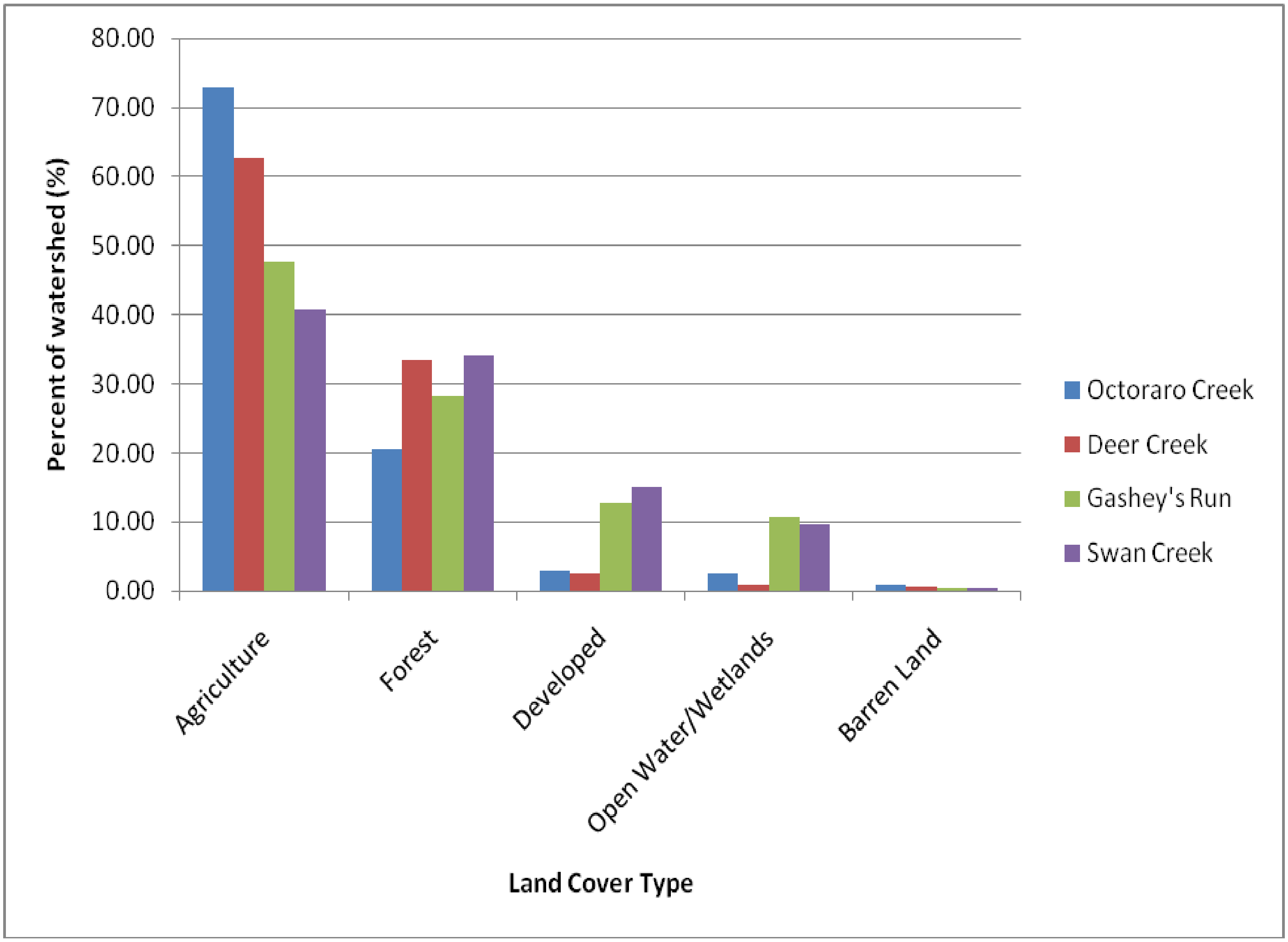

Figure 46: Graph showing the percentages of each land cover type for each watershed.

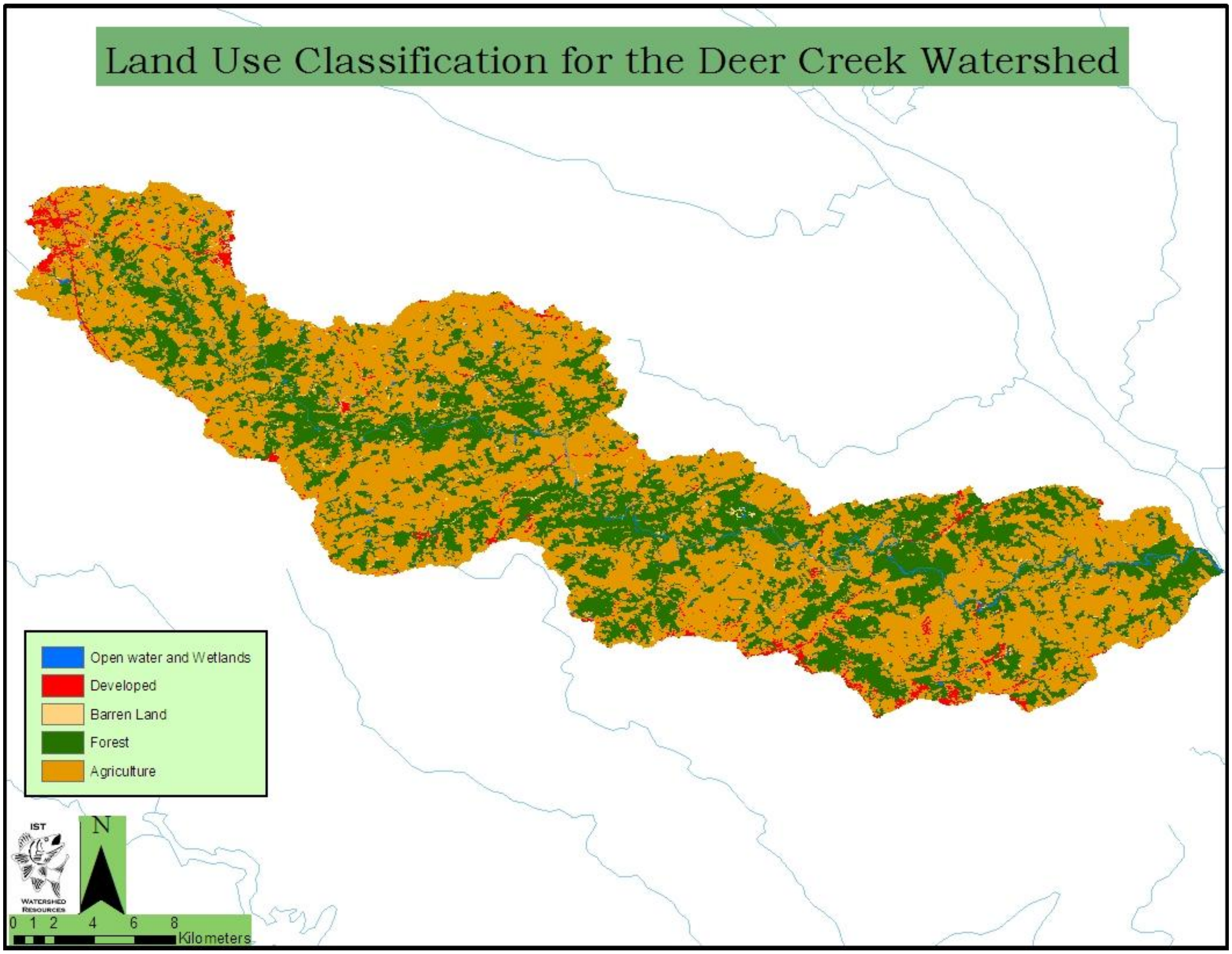

Figure 47: Map showing Deer Creek watershed classified by land cover type.

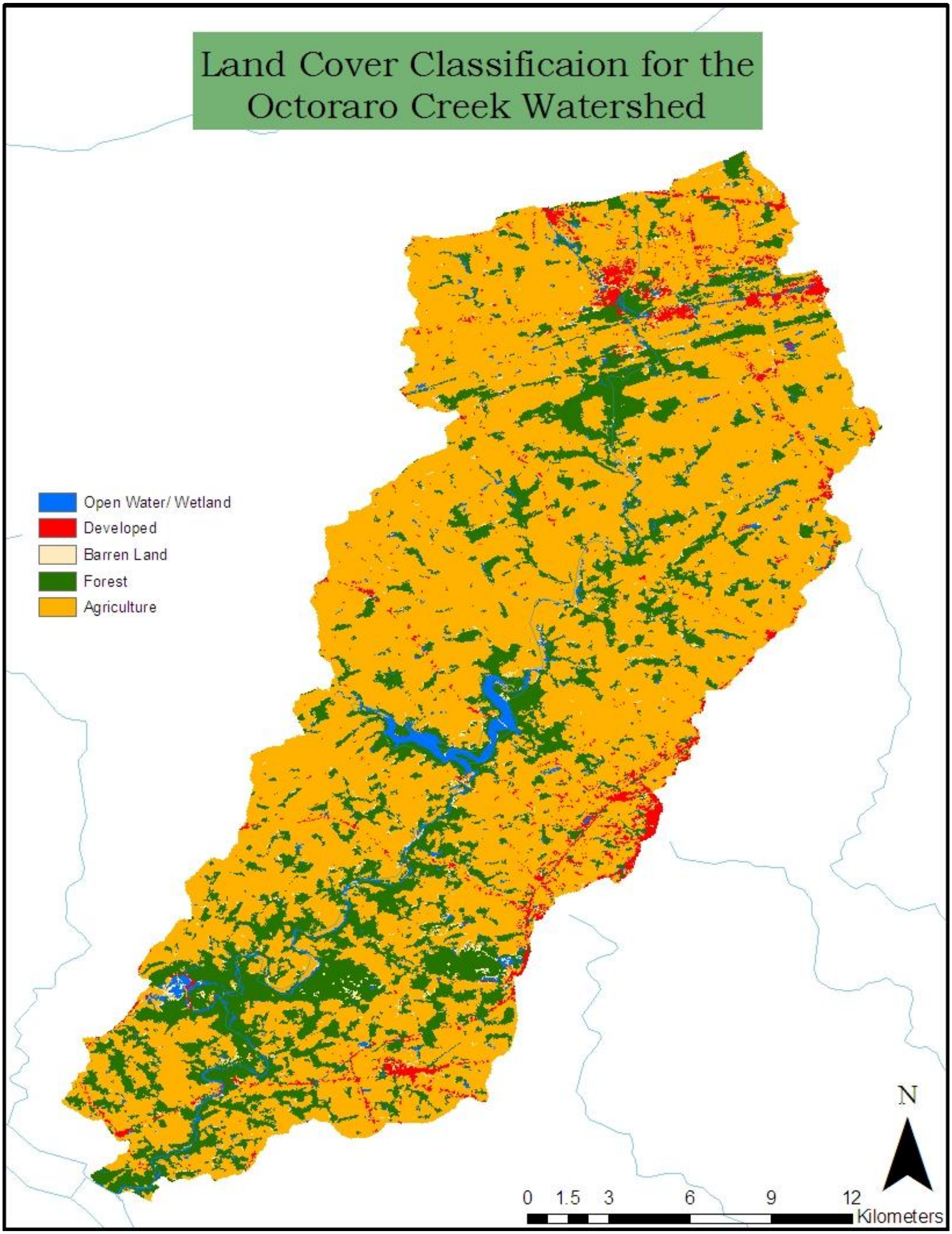

Figure 48: Map showing Octoraro Creek watershed classified by land cover type.

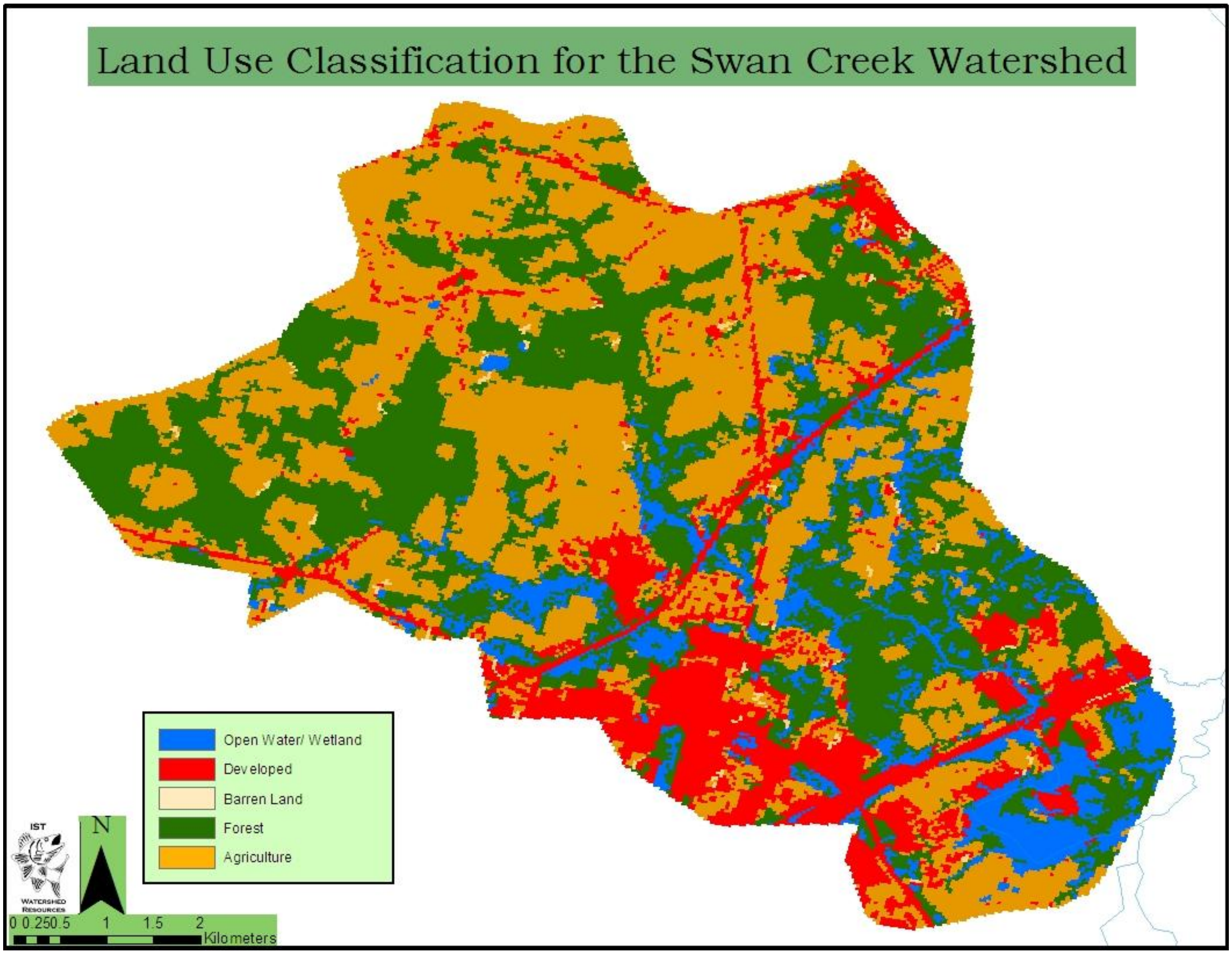

Figure 49: Map showing Swan Creek watershed classified by land cover type.

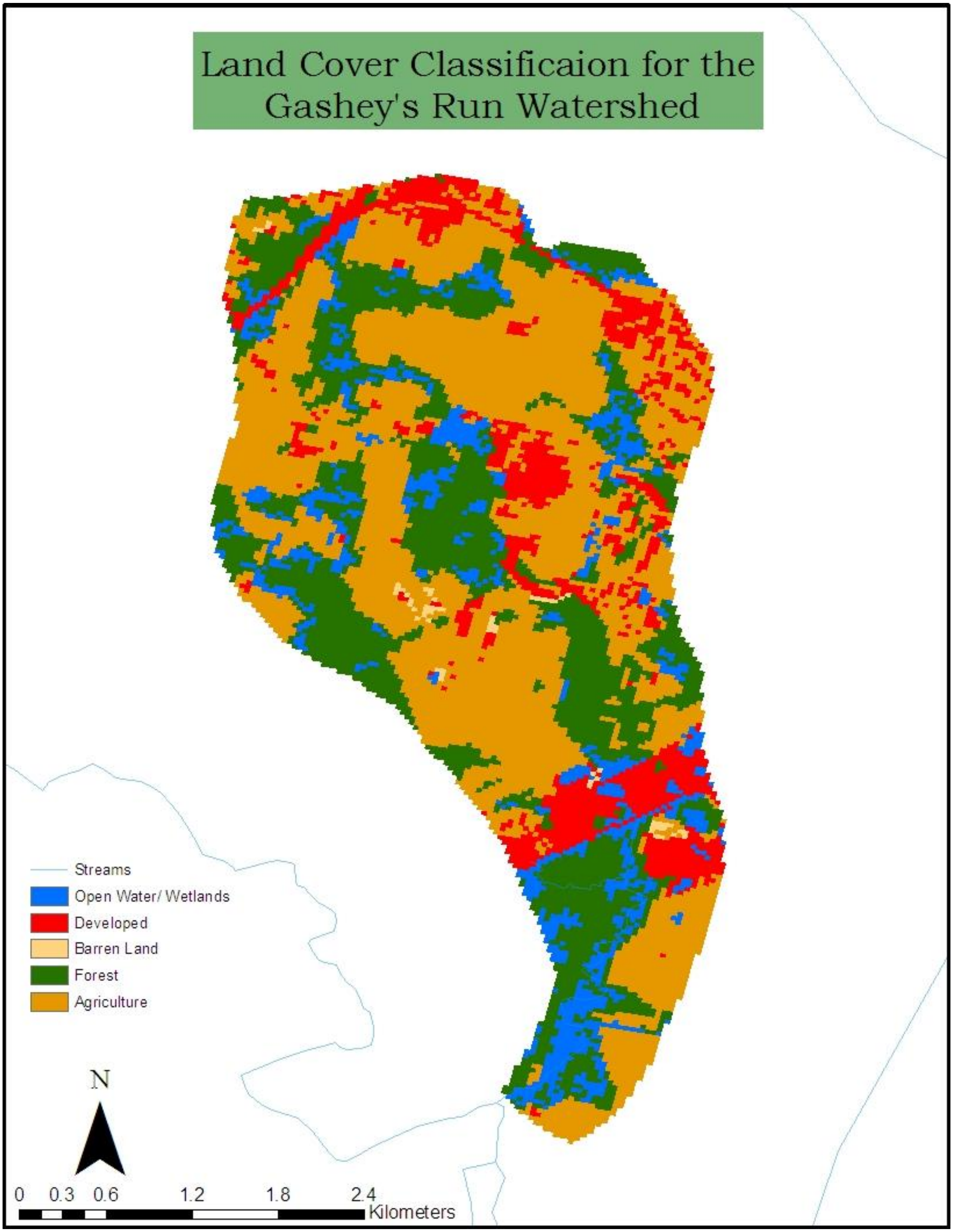

Figure 50: Map showing Gashey’s Run watershed classified by land cover type.

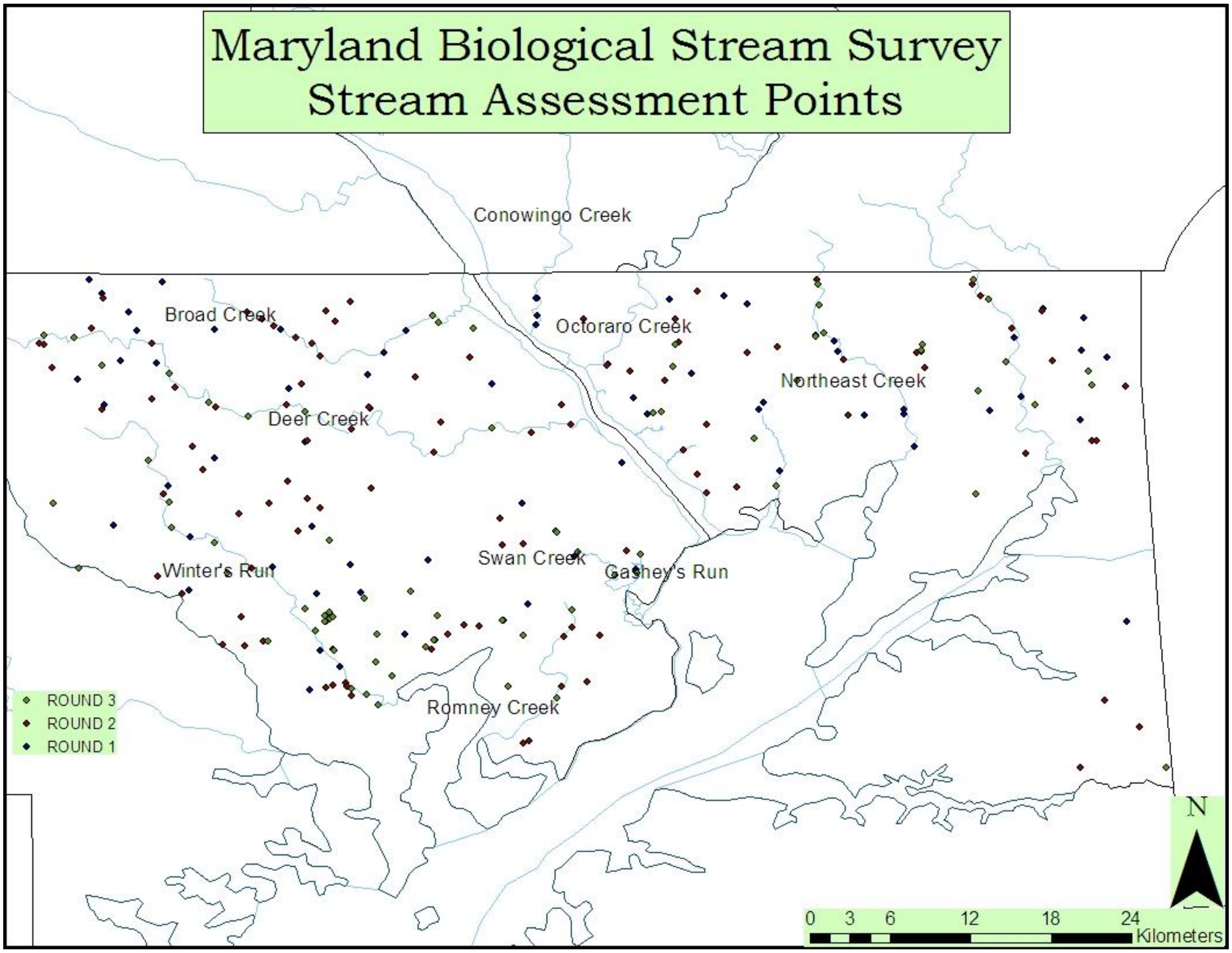

Figure 51: Map showing locations of MBSS stream assessments for all rounds.

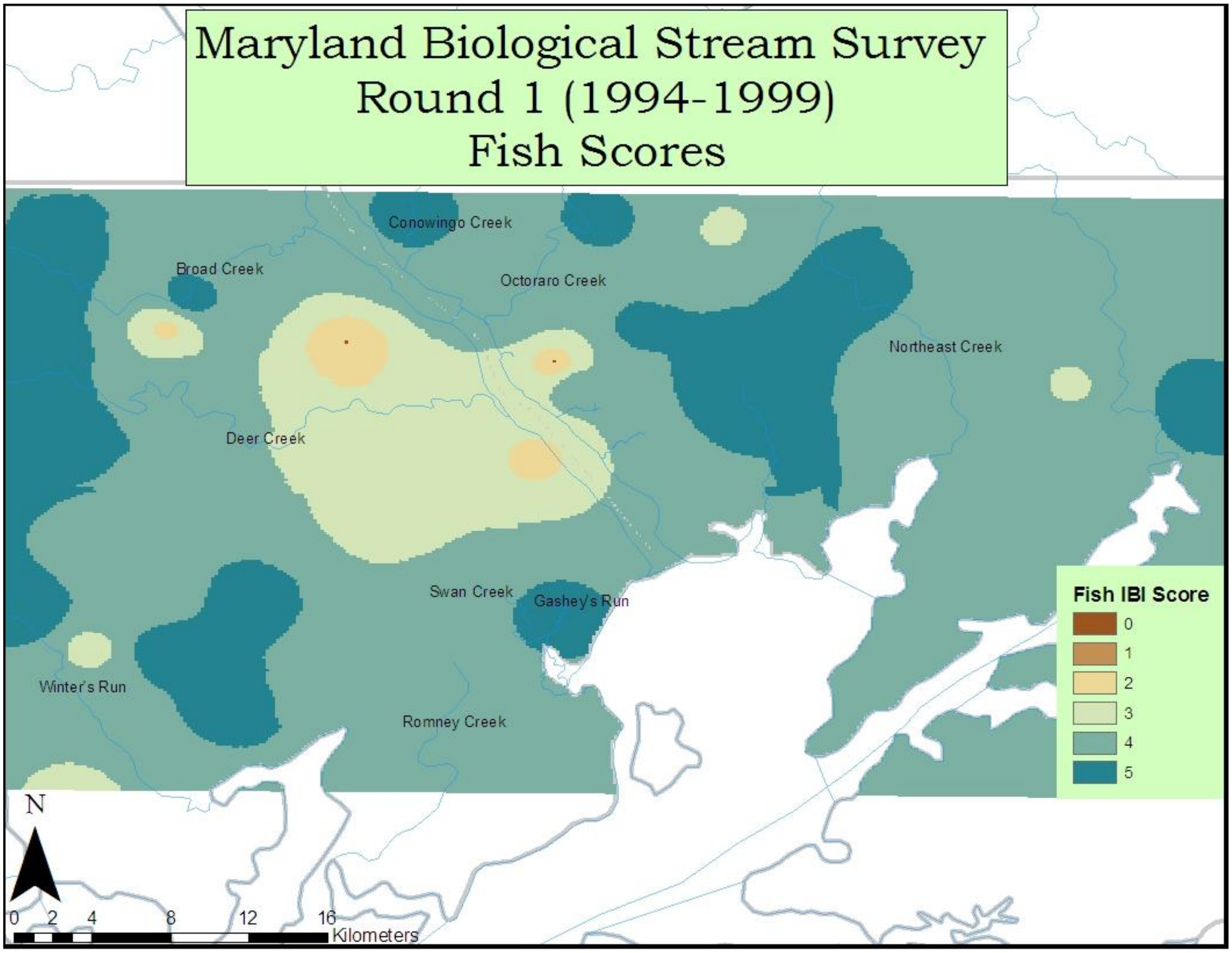

Figure 52: Map showing distribution of fish IBI scores for MBSS Round 1.

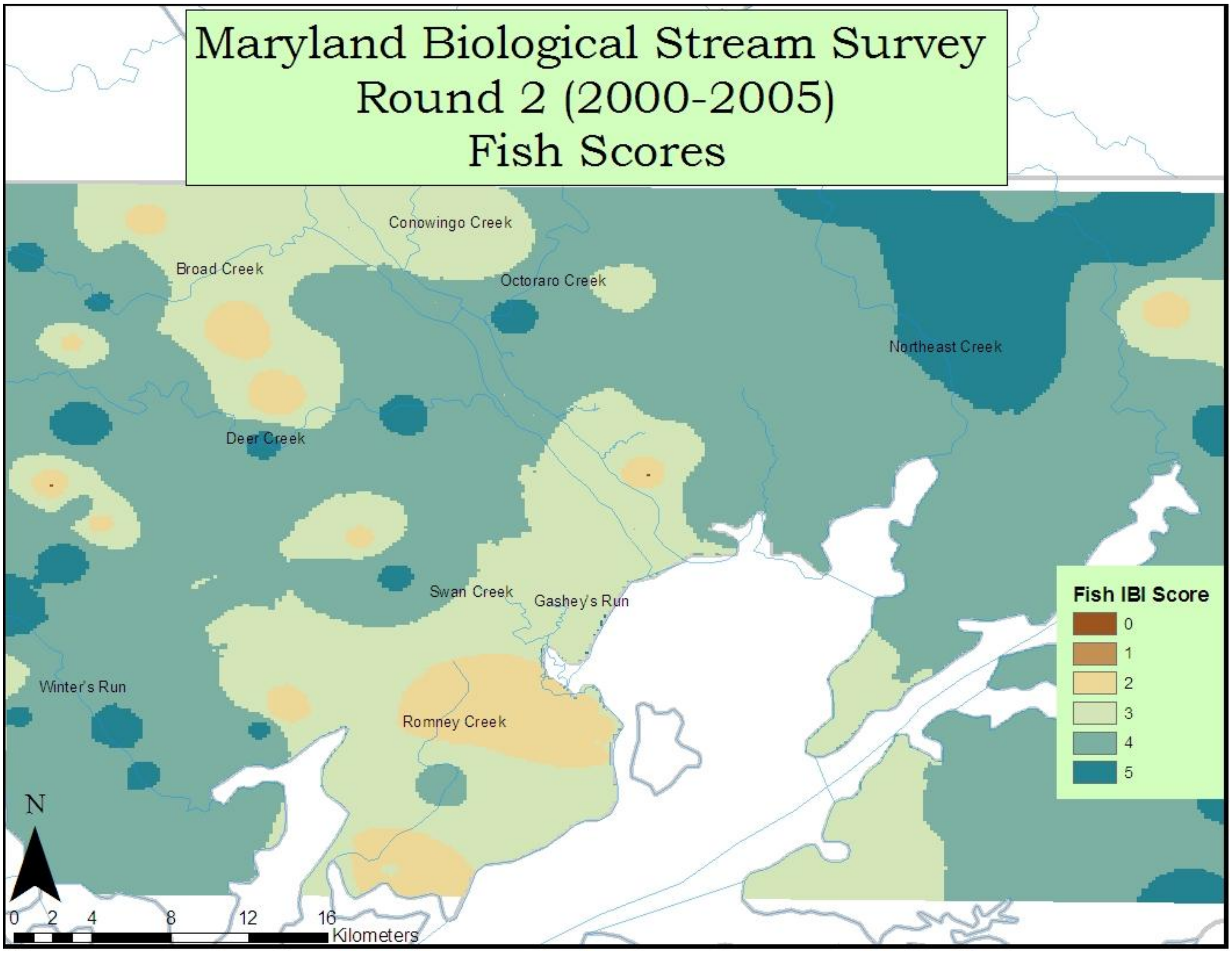

Figure 53: Map showing distribution of fish IBI scores for MBSS Round 2.

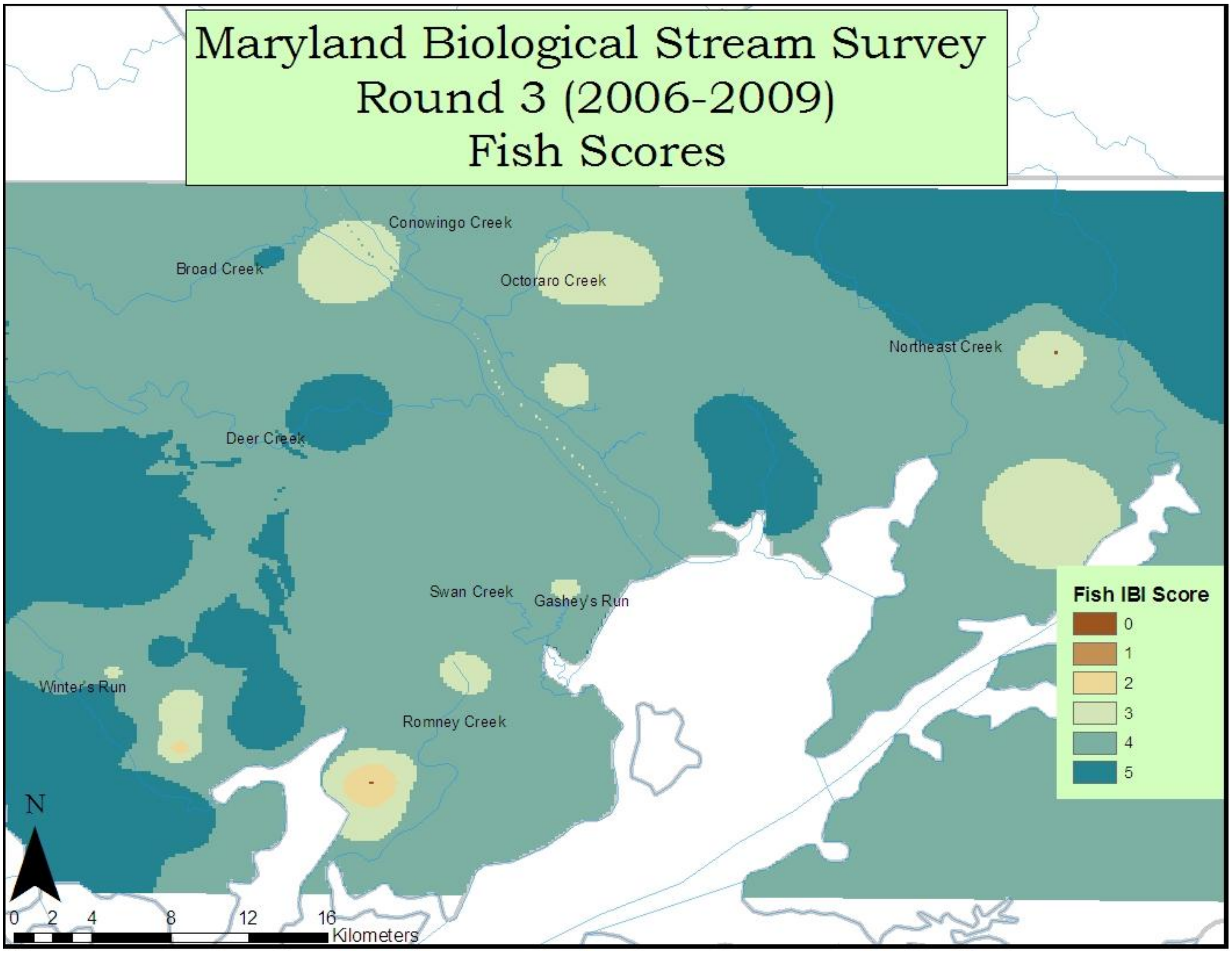

Figure 54: Map showing distribution of fish IBI scores for MBSS Round 3.

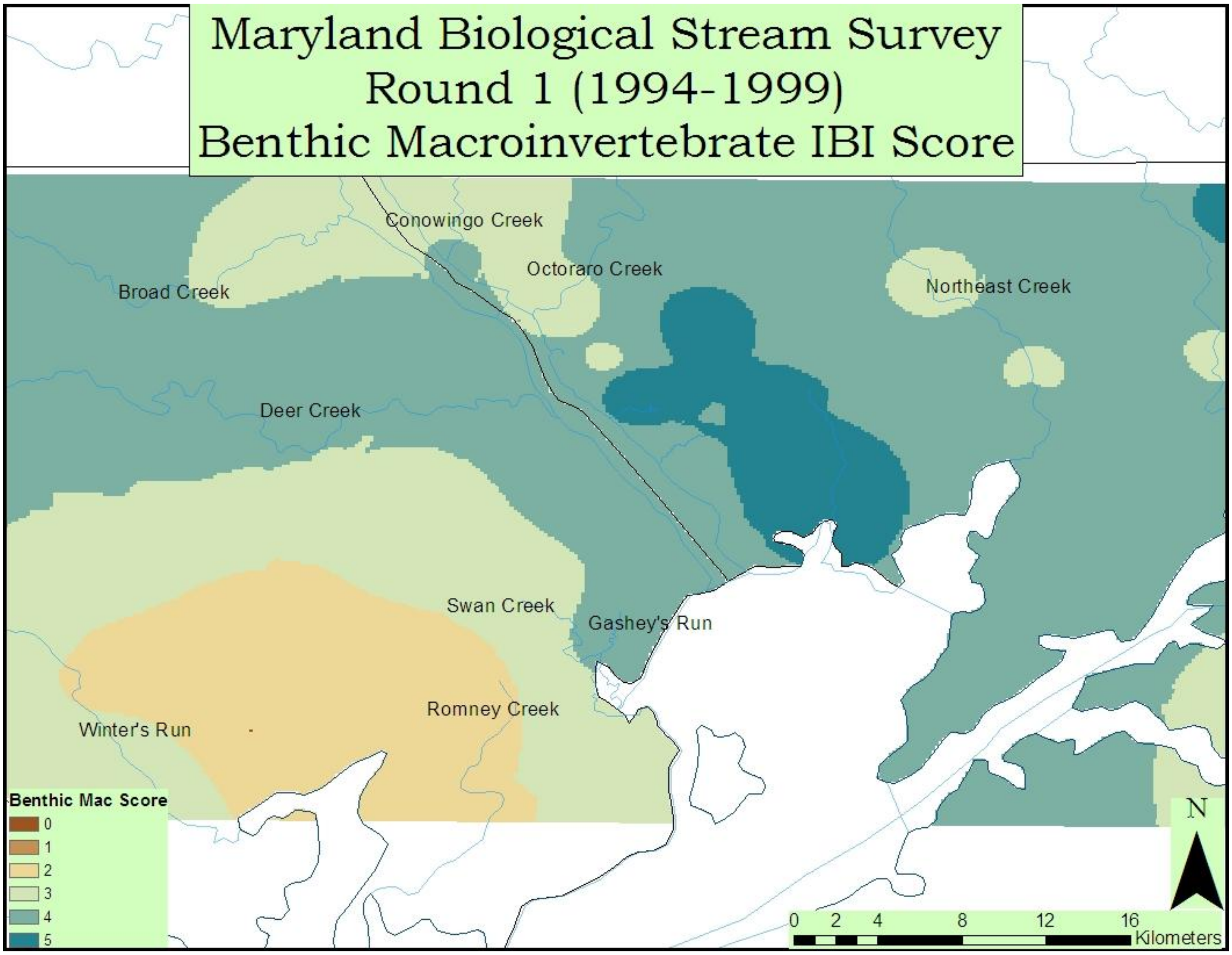

Figure 55: Map showing distribution of benthic macroinvertebrates IBI scores for MBSS Round 1.

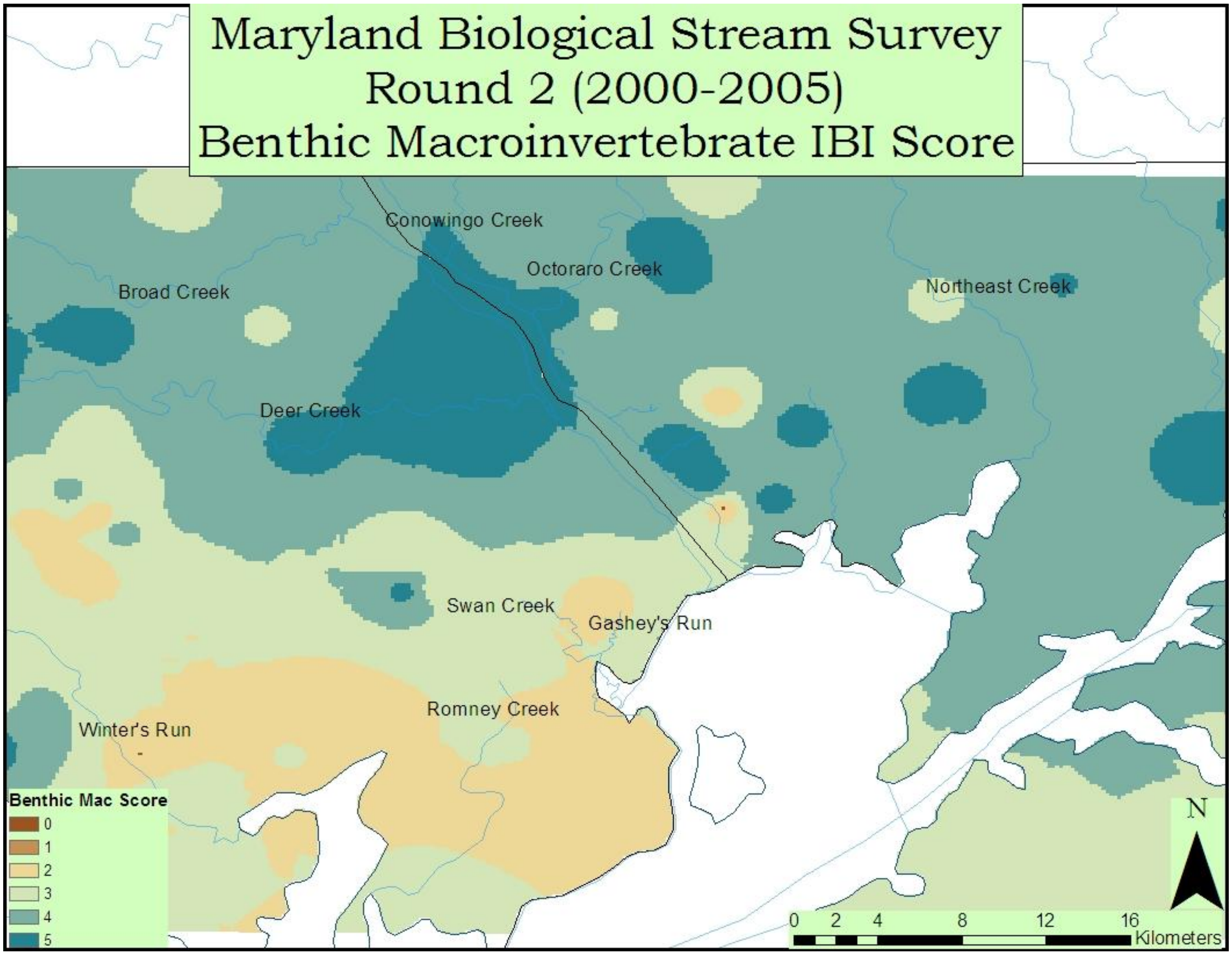

Figure 56: Map showing distribution of benthic macroinvertebrates IBI scores for MBSS Round 2.

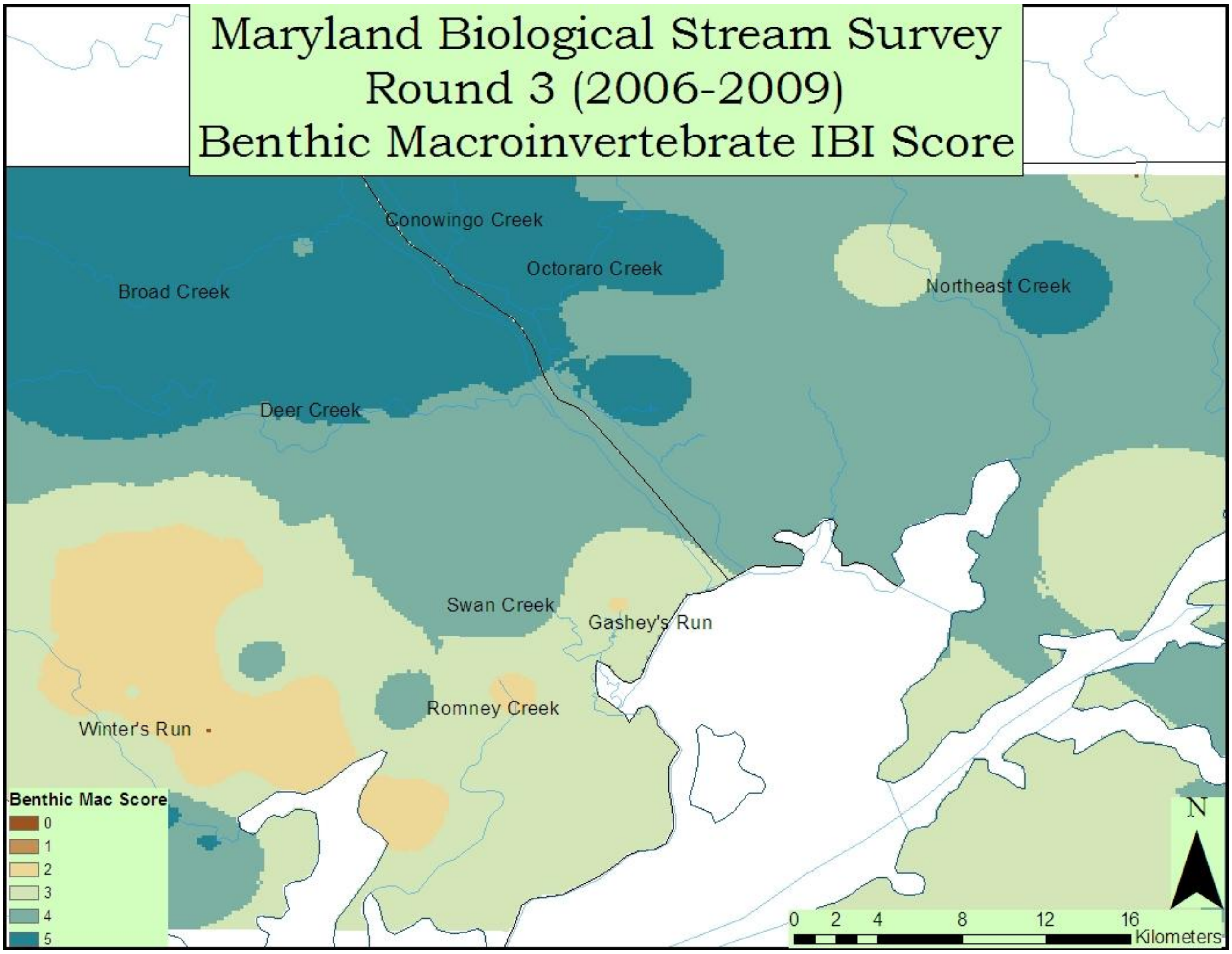

Figure 57: Map showing distribution of benthic macroinvertebrates IBI scores for MBSS Round 3.

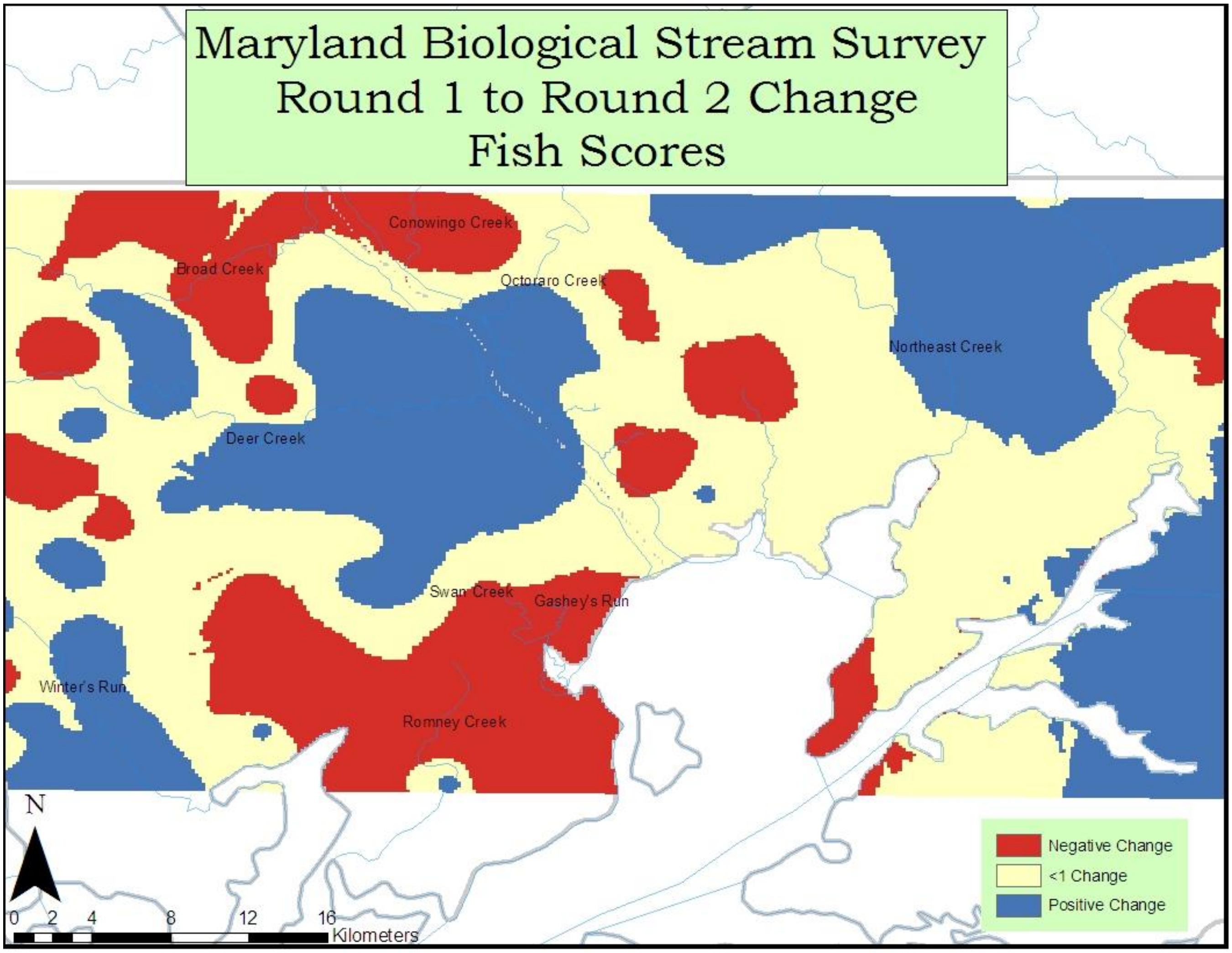

Figure 58: Map showing change in fish IBI scores from MBSS round 1 to round 2.

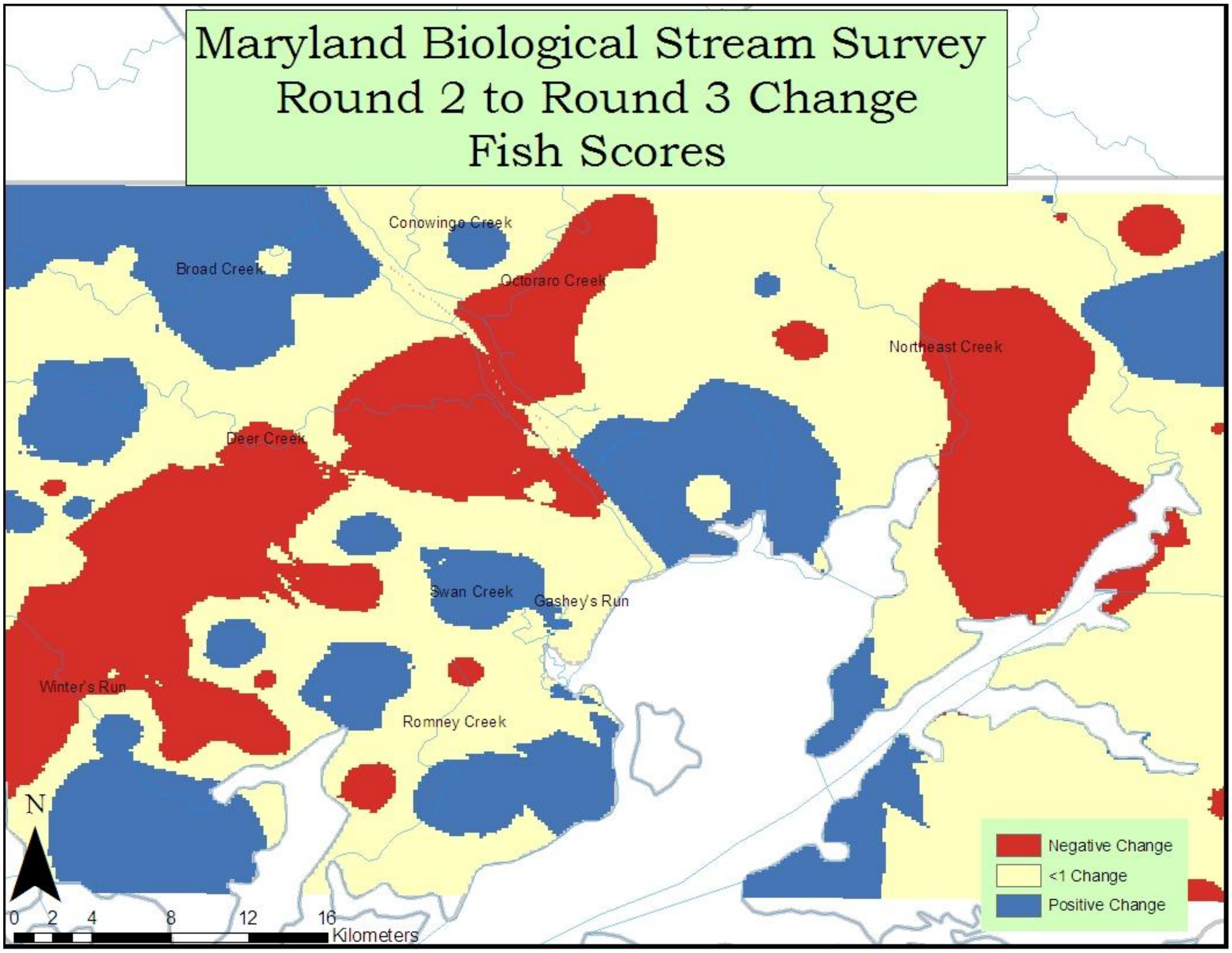

Figure 59: Map showing change in fish IBI scores from MBSS round 2 to round 3.

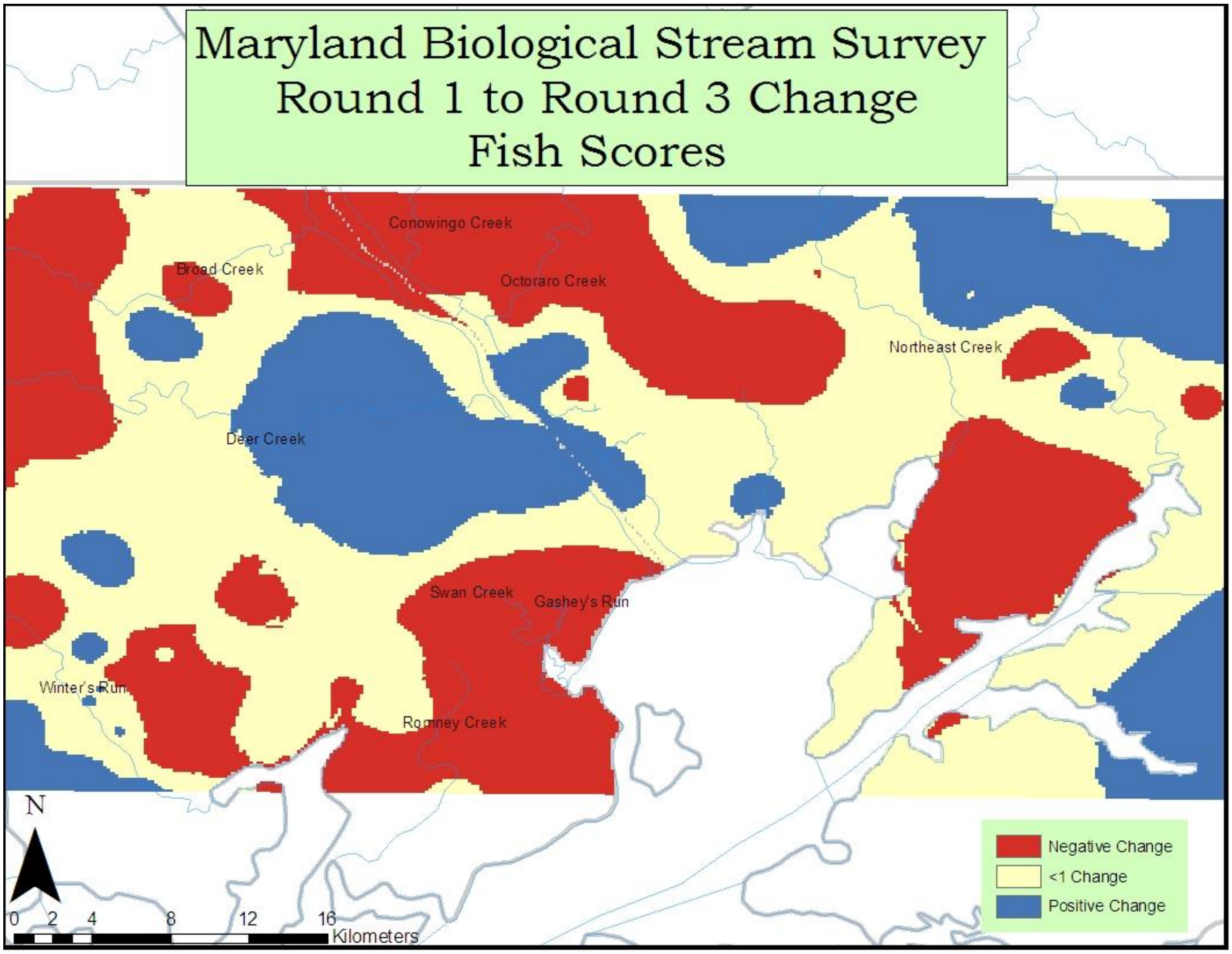

Figure 60: Map showing change in fish IBI scores from MBSS round 1 to round 3.